

Store at
4°C

Cas9 (*S. pyogenes*) (D8Y4K) Rabbit mAb (PE Conjugate)

#54580



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Entrez-Gene ID #901176
UniProt ID #Q99ZW2

New 05/18

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Applications
F
Transfected Only

Species Cross-Reactivity
All

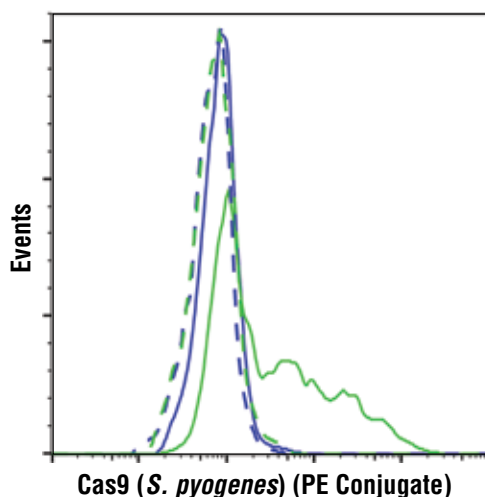
Isotype
Rabbit IgG

Description: This Cell Signaling Technology antibody is conjugated to phycoerythrin (PE) and tested in-house for direct flow cytometric analysis in human cells. This antibody is expected to exhibit the same species cross-reactivity as the unconjugated Cas9 (*S. pyogenes*) (D8Y4K) Rabbit mAb #65832.

Background: The CRISPR associated protein 9 (Cas9) is an RNA-guided DNA nuclease and part of the *Streptococcus pyogenes* CRISPR antiviral immunity system that provides adaptive immunity against extra chromosomal genetic material (1). The CRISPR antiviral mechanism of action involves three steps: (i), acquisition of foreign DNA by host bacterium; (ii), synthesis and maturation of CRISPR RNA (crRNA) followed by the formation of RNA-Cas nuclease protein complexes; and (iii), target interference through recognition of foreign DNA by the complex and its cleavage by Cas nuclease activity (2). The type II CRISPR/Cas antiviral immunity system provides a powerful tool for precise genome editing and has potential for specific gene regulation and therapeutic applications (3). The Cas9 protein and a guide RNA consisting of a fusion between a crRNA and a trans-activating crRNA (tracrRNA) must be introduced or expressed in a cell. A 20-nucleotide sequence at the 5' end of the guide RNA directs Cas9 to a specific DNA target site. As a result, Cas9 can be "programmed" to cut various DNA sites both *in vitro* and in cells and organisms. CRISPR/Cas9 genome editing tools have been used in many organisms, including mouse and human cells (4,5). Research studies demonstrate that CRISPR can be used to generate mutant alleles or reporter genes in rodents and primate embryonic stem cells (6-8).

Specificity/Sensitivity: Cas9 (*S. pyogenes*) (D8Y4K) Rabbit mAb (PE Conjugate) recognizes transfected levels of total Cas9 (*S. pyogenes*) protein. This antibody does not cross-react with Cas9 (*S. aureus*), FnCpf1, and AsCpf1 proteins.

Source/Purification: Monoclonal antibody is produced by immunizing animals with a synthetic peptide corresponding to residues surrounding Val16 of Cas9 (*S. pyogenes*) protein.



Flow cytometric analysis of 293T cells, mock-transfected (blue) or transfected with a construct expressing Cas9 (*S. pyogenes*) (green), using Cas9 (*S. pyogenes*) (D8Y4K) Rabbit mAb (PE Conjugate) (solid lines) or concentration-matched Rabbit (DA1E) mAb IgG XP[®] Isotype Control (PE Conjugate) #5742 (dashed lines).

Storage: Supplied in PBS (pH 7.2), less than 0.1% sodium azide and 2 mg/ml BSA. Store at 4°C. Do not aliquot the antibody. Protect from light. Do not freeze.

Recommended Antibody Dilutions:

Flow Cytometry 1:50

For product specific protocols and a complete listing of recommended companion products please see the product web page at www.cellsignal.com.

Background References:

- (1) Horvath, P. and Barrangou, R. (2010) *Science* 327, 167-70.
- (2) Wiedenheft, B. et al. (2012) *Nature* 482, 331-8.
- (3) Singh, P. et al. (2015) *Genetics* 199, 1-15.
- (4) Cong, L. et al. (2013) *Science* 339, 819-23.
- (5) Mali, P. et al. (2013) *Science* 339, 823-6.
- (6) Li, D. et al. (2013) *Nat Biotechnol* 31, 681-3.
- (7) Shen, B. et al. (2013) *Cell Res* 23, 720-3.
- (8) Niu, Y. et al. (2014) *Cell* 156, 836-43.

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Applications: W—Western IP—Immunoprecipitation IHC—Immunohistochemistry ChIP—Chromatin Immunoprecipitation IF—Immunofluorescence F—Flow cytometry E-P—ELISA-Peptide **Species Cross-Reactivity:** H—human M—mouse R—rat Hm—hamster Mk—monkey Mi—mink C—chicken Dm—D. melanogaster X—Xenopus Z—zebrafish B—bovine Dg—dog Pg—pig Sc—S. cerevisiae Ce—C. elegans Hr—Horse All—all species expected **Species enclosed in parentheses are predicted to react based on 100% homology.**