

Rev01  
Update: Dec,13,2022

**DATASHEET**

# GenCRISPR™ SaCas9 Antibody (11C12), mAb, Mouse

Cat. No.: A01951

## Overview

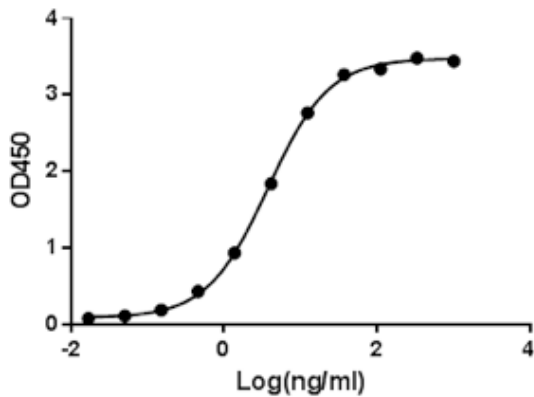
<b>Specificity</b>	The product is specific for Staphylococcus aureus CRISPR/Cas9. This antibody binds with recombinant Staphylococcus aureus CRISPR/Cas9 protein in ELISA and Endogenous overexpressed Staphylococcus aureus CRISPR/Cas9 in immunofluorescence, western blot, immunoprecipitation.
<b>Host Species</b>	Mouse
<b>Immunogen</b>	Recombinant Staphylococcus aureus CRISPR/Cas9

## Properties

<b>Concentration</b>	0.5 mg/ml, lyophilized with PBS, pH 7.4, contains 0.02% sodium azide. (Might be eligible for customized bulk orders.)
<b>Reconstitution</b>	Reconstitute the lyophilized antibody with deionized water (or equivalent) to a final concentration of 0.5 mg/ml.
<b>Purification</b>	Protein A affinity column
<b>Clone ID</b>	11C12

## Examples

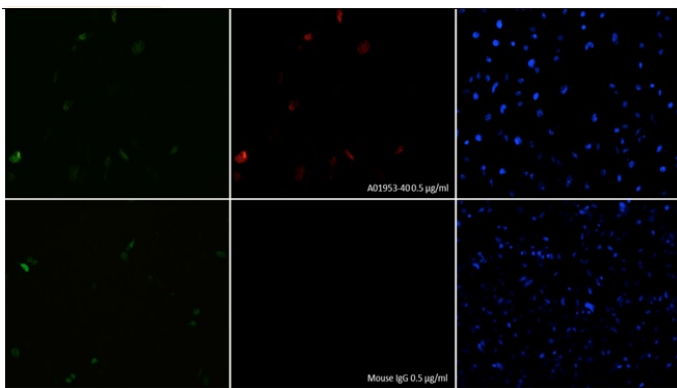
### Clone 11C12 ELISA binding with SaCas9



ELISA binding of GenCRISPR™ SaCas9 Antibody (11C12) (GenScript, A01951-40) with recombinant Staphylococcus aureus CRISPR/Cas9 protein.

Coating antigen: SaCas9, 1 µg/ml.

SaCas9 antibody dilution start from 1000 ng/ml,  $EC_{50}$  = 3.75 ng/ml.

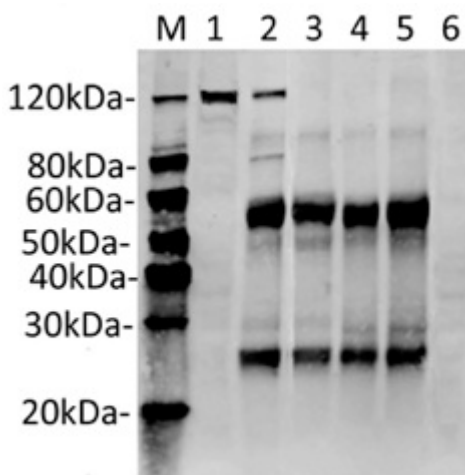


Immunofluorescence staining of GenCRISPR™ SaCas9 Antibody (11C12) (GenScript, A01951-40) with SaCas9 in HeLa cells.

HeLa cells transfected with PX458 (SaCas9(BB)-2A-GFP) (colored green) were fixed with 4% Poly-Formaldehyde (5min) and then blocked in 3% BSA 30min.

The cells were then incubated with GenCRISPR™ SaCas9 Antibody (11C12) (GenScript, A01951-40) at 0.5 µg/ml mouse IgG at 0.5 µg/ml at room temperature for 2h, followed by a further incubation at 37°C for 1h with Goat Anti-Mouse IgG Antibody (H&L) [ifluor 555], pAb (GenScript) (colored red) at 5 µg/ml.

DAPI was used to stain the cell nuclei (colored blue) at a concentration of 0.2 µg/ml for 2h at room temperature.



Immunoprecipitation of SaCas9 from HeLa cell transfected with PX458 (SaCas9(BB)-2A-GFP) by using GenCRISPR™ SaCas9 Antibody (11C12) (GenScript, A01951-40).

Predicted band size: 124 kDa

Loading:

Lane 1: HeLa cell lysate transfected with PX458 (SaCas9(BB)-2A-GFP) 50µg (Input)

Lane 2: Protein A MagBeads (20µl) + Purified antibody of A01951-40 (10µg) + HeLa cell lysate transfected with PX458 (SaCas9(BB)-2A-GFP) (200µg)

Lane 3: Protein A MagBeads (20µl) + Purified antibody of A01951-40 (10µg) + HeLa cell lysate (200µg)

Lane 4: Protein A MagBeads (20µl) + Mouse IgG (10µg) + HeLa cell lysate transfected with PX458 (SaCas9(BB)-2A-GFP) (200µg)

Lane 5: Protein A MagBeads (20µl) + Mouse IgG (10µg) + HeLa Cell lysate (200µg)

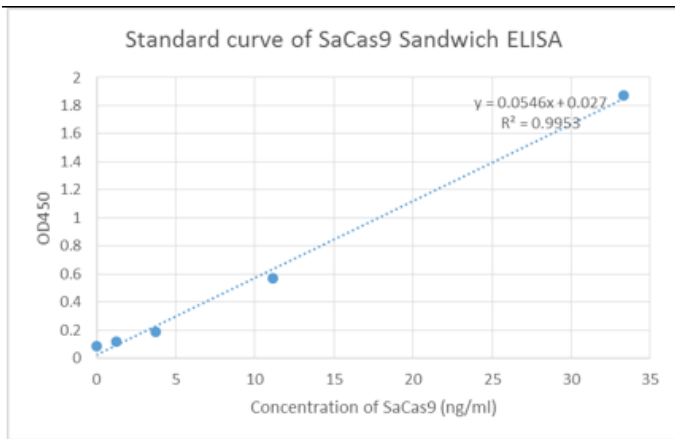
Lane 6: HeLa Cell lysate (50µg)

Primary Antibody:

GenCRISPR™ SaCas9 Antibody (11C12) (GenScript, A01951-40)  
 1 µg/ml

Secondary antibody:

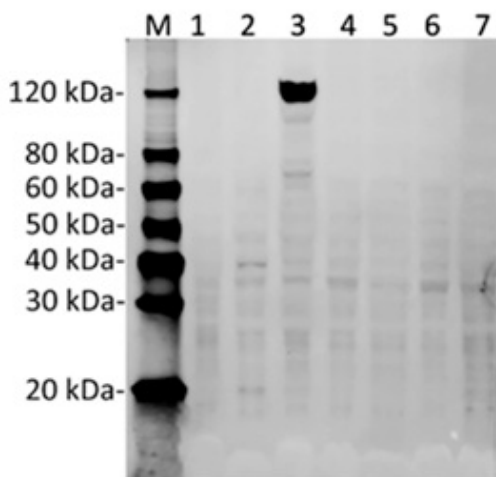
Goat anti-Mouse IgG (H&L) [IRDye<sup>800</sup>], 0.125 µg/ml



Standard curve of SaCas9 Sandwich ELISA. The SaCas9 Sandwich ELISA assay is developed by using GenCRISPR™ SaCas9 Antibody (11C12) (GenScript, A01951-40) and GenCRISPR™ SaCas9 Antibody (26H10) (GenScript, A01952-40) as capture and detection antibody, respectively. These two antibodies recognize different epitopes.

In this ELISA assay, GenCRISPR™ SaCas9 Antibody (26H10) (GenScript, A01952-40) was labeled with Biotin. GenScript can provide customized conjugation service for this product per customer's request.

The sensitivity is <1 ng/ml and the detection range is 0-30 ng/ml.



Western Blot analysis of HeLa transfected with various

plasmids with GenCRISPR™ SaCas9 Antibody (11C12) (GenScript, A01951-40). The different HeLa transfected with various plasmids indicate the minimum cross reaction of the antibody.

Predicted band size: 124 kDa

Loading:

Lane 1: 50µg HeLa cell lysate transfected with StCas9(BB)-2A-GFP (G3ECR1, *Streptococcus thermophilus*)

Lane 2: 50µg HeLa cell lysate transfected with pSpCas9(BB)-2A-GFP (PX458, Q99ZW2, *Streptococcus pyogenes* serotype M1)

Lane 3: 50µg HeLa cell lysate transfected with SaCas9(BB)-2A-GFP (J7RUA5, *Staphylococcus aureus*)

Lane 4: 50µg HeLa cell lysate transfected with AsCpf1(BB)-2A-GFP (U2UMQ6, *Acidaminococcus* sp. (strain BV3L6))

Lane 5: 50µg HeLa cell lysate transfected with FnCpf1(BB)-2A-GFP (A0Q7Q2, *Francisella tularensis* subsp. *novicida* (strain U112))

Lane 6: 50µg HeLa cell lysate transfected with LbCpf1(BB)-2A-GFP (A0A182DWE3, *Lachnospiraceae* bacterium ND2006)

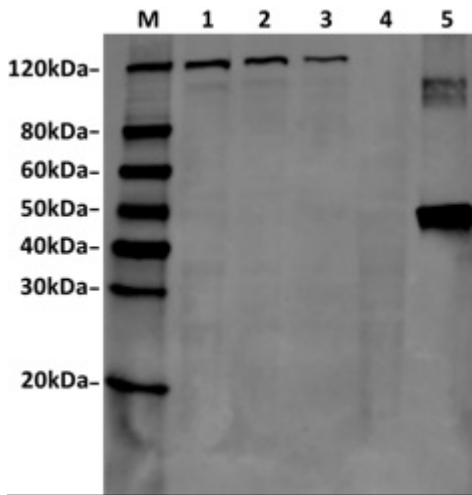
Lane 7: 50µg HeLa cell lysate (Non-transfected)

Primary Antibody:

GenCRISPR™ SaCas9 Antibody (11C12) (GenScript, A01951-40)  
 1 µg/ml

Secondary antibody:

Goat anti-Mouse IgG (H&L) [IRDye<sup>800</sup>], 0.125 µg/ml



Western Blot of HeLa transfected with PX458 (SaCas9(BB)-2A-GFP) or untransfected cell lysates with GenCRISPR™ SaCas9 Antibody (11C12) (GenScript, A01951-40). The different concentration of cell lysates indicates the high affinity and sensitivity of the antibody.

Predicted band size: 124 kDa

Predicted band size of recombinant protein: 47.5 kDa

Loading:

Lane 1: 50 µg HeLa transfected with SaCas9(BB)-2A-GFP cell Lysate

Lane 2: 25 µg HeLa transfected with SaCas9(BB)-2A-GFP cell Lysate

Lane 3: 10 µg HeLa transfected with SaCas9(BB)-2A-GFP cell Lysate

Lane 4: 50 µg Untransfected HeLa cell Lysate

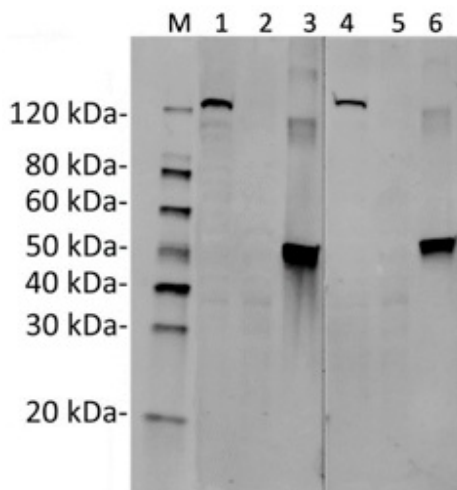
Lane 5: 40 ng SaCas9 recombinant protein

Primary Antibody:

GenCRISPR™ SaCas9 Antibody (11C12) (GenScript, A01951-40)  
 1 µg/ml

Secondary Antibody:

Goat anti-Mouse IgG (H&L) [IRDye<sup>800</sup>], 0.125 µg/ml



Western Blot of HeLa transfected with PX458 (SaCas9(BB)-2A-GFP) or untransfected cell lysates with two independent antibodies: GenCRISPR™ SaCas9 Antibody (11C12) (GenScript, A01951-40) and GenCRISPR™ SaCas9 Antibody (26H10) (GenScript, A01952-40). The correlated pattern indicates the high specificity of these two antibodies.

Predicted band size: 124 kDa

Predicted band size of recombinant protein: 47.5 kDa

Loading:

Lane 1: 50 µg HeLa transfected with SaCas9(BB)-2A-GFP cell Lysate

Lane 2: 50 µg Untransfected HeLa cell Lysate

Lane 3: 40 ng SaCas9 recombinant protein

Lane 4: 50 µg HeLa transfected with SaCas9(BB)-2A-GFP cell Lysate

Lane 5: 50 µg Untransfected HeLa cell Lysate

Lane 6: 40 ng SaCas9 recombinant protein

Primary Antibody:

Lane 1~3: GenCRISPR™ SaCas9 Antibody (11C12) (GenScript, A01951-40) 1 µg/ml

Lane 4~6: GenCRISPR™ SaCas9 Antibody (26H10) (GenScript,

A01952-40) 1 µg/ml

Secondary Antibody:

Goat anti-Mouse IgG (H&L) [IRDye<sup>800</sup>] , 0.125 µg/ml

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## Background

**Target Background :** Clustered regularly interspaced short palindromic repeats (CRISPR)/CRISPR-associated (Cas) protein 9 system provides a robust and multiplexable genome editing tool, enabling researchers to precisely manipulate specific genomic elements, and facilitating the elucidation of target gene function in biology and diseases. CRISPR/Cas9 genome editing allows for double-stranded DNA breaks at specific sequences to efficiently disrupt, excise, mutate, insert, or replace genes. The best characterized CRISPR-associated nucleases are the Cas9 proteins from *Streptococcus pyogenes* and *Staphylococcus aureus*. It is important that the precision of transfection and the level of Cas9 expression should be controlled strictly during the editing processes by using specific anti-CRISPR/Cas9 antibodies.

**Synonyms :** Mouse monoclonal to *Staphylococcus aureus* Cas9/SaCas9