Cas9 Nuclease (GMP grade) – DMF Filed

Product Information

Source: Recombinant *S. pyogenes* CRISPR-Cas9 Protein (GMP grade) is expressed from *E.coli* for genome editing applications with CRISPR technology. It contains Asp2-Asp1368.

Molecular Weight: 163kDa

Formulation: Supplied as 0.22 μm filtered solution in 30 mM Tris-HCl, 0.3 M NaCl, 50% Glycerol, 0.1 mM EDTA, pH7.4. Please dilute to the desired concentration according to the concentration of the solution shown on the product label.

Transportation: This product is supplied and shipped as sterile liquid solution with dry ice.

Storage: -20°C

Size: 3mg/tube

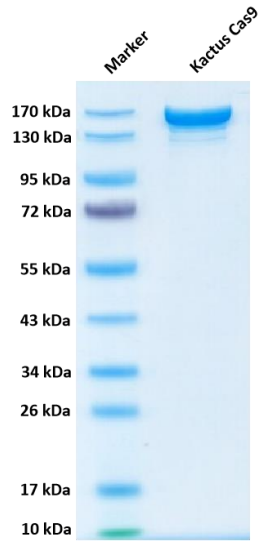
Quality Requirement

Item	Acceptable Standards
Concentrate	9.5-12.5mg/ml
Purity	≥95%(Bis-Tris)
	≥95%(RP-HPLC)
	≥95%(SEC-HPLC)
Activity	In vitro cleavage activity > 85%
Endotoxin	≤10EU/mg
Residual Host Protein	≤100ng/ml
Residual Host Cell DNA	≤200ng/ml
Sterility	Negative
Mycoplasma	Negative

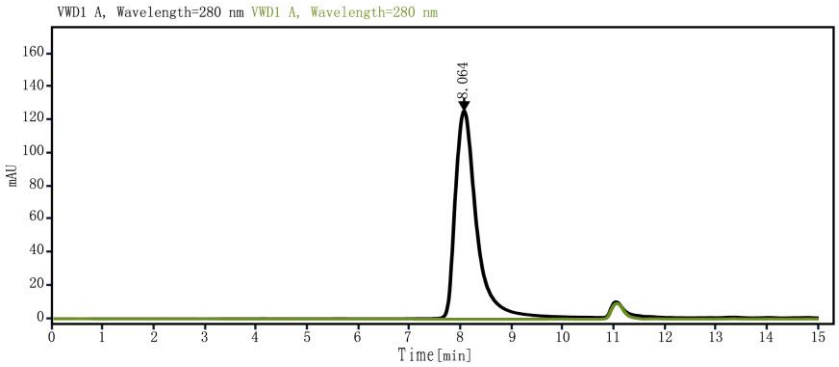
See certificate of analysis for more details.

Assay Data

Tris-Bis PAGE

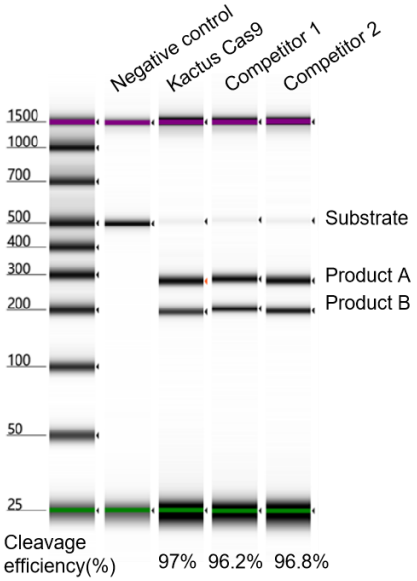


SEC-HPLC



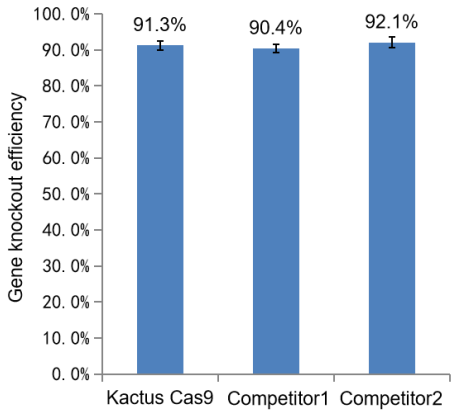
The purity of Kactus Cas9 detected by Tris-Bis-PAGE and SEC-HPLC is higher than 95%.

In vitro cleavage activity



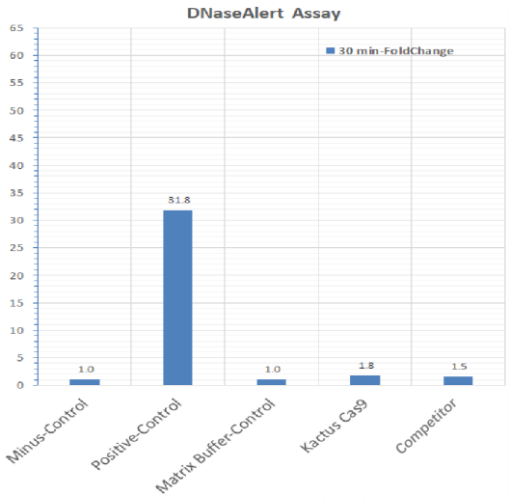
Cas9 cuts the substrate DNA standard through the in vitro cutting experiment. The result of the figure shows that the cleavage activity of Kactus Cas9 is equivalent to that of other brands.

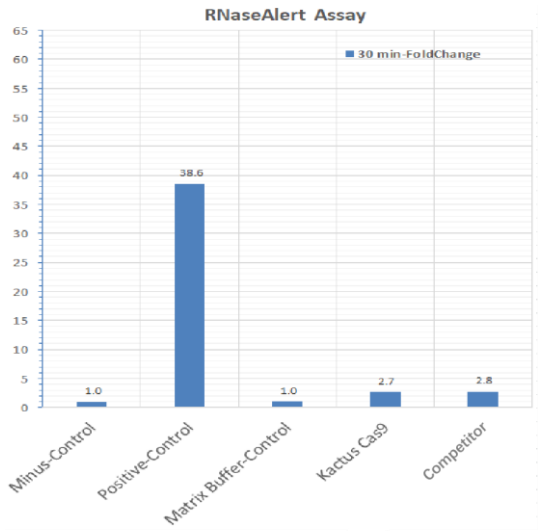
Gene knockout efficiency in cell line



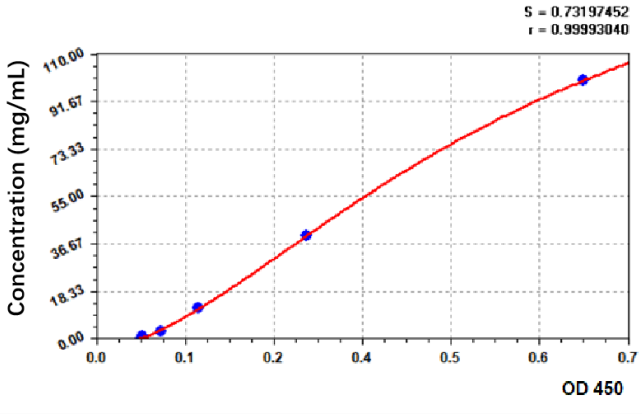
Cas9 is electrotransfected into 293T cell line in the form of RNP to knock out the target gene. The results show that Kactus Cas9 has the same knockout efficiency in cells as foreign brand.

Residual DNase and RNase





Test samples which have 3-fold more fluorescence than the minus-DNase/RNase control should be considered contaminated with DNase/RNase，and the test result of Kactus Cas9 was negative.



ELISA assay is used for determining the residual host-cell proteins and the result is 3.0 ng/ml, which is far lower than the detection standard (<100 ng/ml).

Background

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). In type II CRISPR systems correct processing of pre-crRNA requires a trans-encoded small RNA (tracrRNA), endogenous ribonuclease 3 (rnc) and Cas9.