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DATASHEET

GenCRISPR™ Ultra NLS-Cas9-Research

Cat. No.: Z03621-0.5; Z03621-1; Z03621-5 Size: 0.5 mg / 1 mg / 5 mg

Product Introduction

The GenCRISPR[™] Ultra Cas9 product line provides customers with a selection of research use, basic GMP and GMP compliant Cas9 nucleases. The Cas9 protein can be formed with the guide RNA into a ribonucleoprotien (RNP) complex. The use of an RNP complex to perform gene editing has been shown to reduce the challenges encountered with other CRISPR gene editing techniques such as viral and plasmid delivery. Challenges include off-target effects, cell viability and trascprition/translational challenges.

GenCRISPR[™] Ultra Cas9 is produced by expression in an E. *coli* strain carrying a plasmid encoding the Cas9 gene from *Streptococcus pyogenes* with an N-terminal nuclear localization signal (NLS).

Source: Recombinant Cas9 with an N-terminal NLS expressed by E.*coli* Species: S. *pyogenes* Tag: Tag-free Molecular Weight: ~160 kDa Concentration: 10 mg/mL Active temperature: This Cas9 is active at 37 °C. **Formulation:** Supplied as a solution of 25 mM Tris, 300 mM NaCl, 0.1 mM EDTA, 50% Glycerol pH 8.0 at 25°C.

Storage& Stability: This product remains stable up to 12 months at -20°C. Avoid repeated freeze-thaw cycles.

Application: gRNA-dependent double-stranded DNA cleavage

Assay	Specifications	
Appearance	Clear, colorless liquid	
Purity	≥ 95% as analyzed by SDS-PAGE ≥ 90% as analyzed by SEC-HPLC	
Concentration by A280	10 mg/ml ± 1 mg/ml	
Bioactivity (in vitro)	≥ 90%	
Residual DNase	Non-specific DNase activity	
Residual RNase	Non-specific RNase activity	

Quality Control Specifications

GenScript USA, Inc. 860 Centennial Ave. Piscataway, NJ 08854



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Endotoxin Level		≤ 10 EU/mg as analyzed by gel clotting method			
Data Images		Cas9)+gRNA		_
+Cas9 (ng)	0	0	50	50	
3000 2000 1500	-	-			Uncut DNA

DNA fragments

A 20 µl reaction in 1 × Cas9 Nuclease Reaction Buffer containing 160 ng linearized plasmid, 100 ng gRNA, and 50 ng GenCRISPR™ Ultra Cas9 for 2 hours at 37°C results in a digestion efficiency of linearized plasmid higher than 90%, as determined by agarose gel electrophoresis.

Key Features

High knockout efficiencies: Consistent high editing efficiency in *in-vitro* and *in-vivo*.
Tag-free: Amino acid is free from additional tagging amino acid.
Animal-free: Plant-based culture media, enzyme-free chromatographic purification.
DNA-free: No external DNA added to the system.

References

- 1. Jinek, Martin, et al. "A programmable dual-RNA–guided DNA endonuclease in adaptive bacterial immunity." *science* 337.6096 (2012): 816-821.
- 2. Chen, Sean, et al. "Highly efficient mouse genome editing by CRISPR ribonucleoprotein electroporation of zygotes." *Journal of Biological Chemistry* 291.28 (2016): 14457-14467.
- 3. Larson, Matthew H., et al. "CRISPR interference (CRISPRi) for sequence-specific control of gene expression." *Nature protocols* 8.11 (2013): 2180-2196.
- Ran, F. Ann, et al. "Genome engineering using the CRISPR-Cas9 system." *Nature protocols* 8.11 (2013): 2281-2308.
- 5. Kim, Sojung, et al. "Highly efficient RNA-guided genome editing in human cells via delivery of purified Cas9 ribonucleoproteins." *Genome research* 24.6 (2014): 1012-1019.

For laboratory research use only. Direct human use, including taking orally and injection and clinical use are forbidden.