

New England Biolabs Certificate of Analysis

Product Name: Cas9 Nuclease, *S. pyogenes*
Catalog Number: M0386T
Concentration: 20 μ M
Packaging Lot Number: 10062630
Expiration Date: 10/2021
Storage Temperature: -20°C
Storage Conditions: 10 mM Tris-HCl, 300 mM NaCl, 1 mM DTT, 0.1 mM EDTA, 50 % Glycerol, (pH 7.4 @ 25°C)
Specification Version: PS-M0386T/M v1.0

Cas9 Nuclease, <i>S. pyogenes</i> Component List			
NEB Part Number	Component Description	Lot Number	Individual QC Result
M0386TVIAL	Cas9 Nuclease, <i>S. pyogenes</i>	10057220	Pass
B7203SVIAL	NEBuffer™ 3.1	10053974	Pass

Assay Name/Specification	Lot # 10062630
Non-Specific DNase Activity (16 Hour) A 50 μ l reaction in NEBuffer 3.1 containing 1 μ g of Lambda DNA and a minimum of 1 pmol of Cas9 Nuclease, <i>S. pyogenes</i> incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.	Pass
Protein Purity Assay (SDS-PAGE) Cas9 Nuclease, <i>S. pyogenes</i> is \geq 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.	Pass
RNase Activity (Extended Digestion) A 10 μ l reaction in NEBuffer 4 containing 40 ng of a 300 base single-stranded RNA and a minimum of 1 pmol of Cas9 Nuclease, <i>S. pyogenes</i> is incubated at 37°C. After incubation for 16 hours, >90% of the substrate RNA remains intact as determined by gel electrophoresis using fluorescent detection.	Pass
Functional Testing (Targeted Digestion) A 20 μ l reaction in NEBuffer 3.1 containing 20 nM of 100 bp FAM and ROX-labeled double-stranded target DNA, 100 nM sgRNA, and 100 nM Cas9 Nuclease, <i>S. pyogenes</i> incubated for 1 hour at 37°C results in \geq 90% targeted digestion of the substrate DNA as determined by capillary electrophoresis.	Pass

Assay Name/Specification	Lot # 10062630
<p>Endonuclease Activity (Nicking) A 50 µl reaction in NEBuffer 3.1 containing 1 µg of supercoiled PhiX174 DNA and a minimum of 1 pmol of Cas9 Nuclease, <i>S. pyogenes</i> incubated for 4 hours at 37°C results in <10% conversion to the nicked form as determined by agarose gel electrophoresis.</p>	Pass
<p>Exonuclease Activity (Radioactivity Release) A 50 µl reaction in NEBuffer 3.1 containing 1 µg of a mixture of single and double-stranded [³H] <i>E. coli</i> DNA and a minimum of 1 pmol of Cas9 Nuclease, <i>S. pyogenes</i> incubated for 4 hours at 37°C releases <0.1% of the total radioactivity.</p>	Pass

This product has been tested and shown to be in compliance with all specifications.



Jessica Cane
Production Scientist
18 Oct 2019



Jay Minichiello
Packaging Quality Control Inspector
04 Mar 2020