

New England Biolabs Certificate of Analysis

Product Name: EnGen® Sau Cas9
Catalog Number: M0654T
Concentration: 20 µM
Packaging Lot Number: 10129877
Expiration Date: 11/2023
Storage Temperature: -20°C
Storage Conditions: 20 mM Tris-HCl, 300 mM NaCl, 0.1 mM TCEP, 50% Glycerol, (pH 7.5 @ 25°)
Specification Version: PS-M0654T v1.0

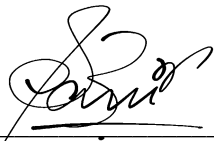
EnGen® Sau Cas9 Component List			
NEB Part Number	Component Description	Lot Number	Individual QC Result
M0654TVIAL	EnGen® Sau Cas9	10128362	Pass
B6003SVIAL	NEBuffer™ r3.1	10116057	Pass

Assay Name/Specification	Lot # 10129877
Exonuclease Activity (Radioactivity Release) A 50 µl reaction in NEBuffer 3.1 containing 1 µg of a mixture of single and double-stranded [³ H] E. coli DNA and a minimum of 1 pmol of EnGen® Sau Cas9 incubated for 4 hours at 37°C releases <0.1% of the total radioactivity.	Pass
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 EnGen® Sau Cas9 incubated for 4 hours at 37°C results in <10% conversion to the nicked form as determined by agarose gel electrophoresis.	Pass
RNase Activity (Extended Digestion) A 10 µl reaction in NEBuffer 4 containing 40 ng of a 300 base single-stranded RNA and a minimum of 1 pmol of EnGen® Sau Cas9 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
Protein Purity Assay (SDS-PAGE) EnGen® Sau Cas9 is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.	Pass
Functional Testing (Targeted Digestion)	Pass

Assay Name/Specification	Lot # 10129877
<p>A 20 µl reaction in NEBuffer 3.1 containing 20 nM of 515 bp FAM and ROX-labeled double-stranded target DNA, 100 nM sgRNA, and 100 nM EnGen® Sau Cas9 incubated for 15 minutes at 37°C results in ≥90% targeted digestion of the substrate DNA as determined by capillary electrophoresis.</p> <p>Non-Specific DNase Activity (16 Hour) A 50 µl reaction in NEBuffer 3.1 containing 1 µg of Lambda DNA and a minimum of 1 pmol of EnGen® Sau Cas9 incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.</p>	<p>Pass</p>

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

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Bhairavi Jani
Production Scientist
02 Dec 2021



Michael Tonello
Packaging Quality Control Inspector
02 Dec 2021