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## New England Biolabs Certificate of Analysis

Product Name: EnGen® Sau Cas9

Catalog Number: M0654SConcentration:  $1 \mu M$ Lot Number: 10040102Expiration Date: 03/2021Storage Temperature:  $-20^{\circ}C$ 

Storage Conditions: 20 mM Tris-HCl, 300 mM NaCl, 0.1 mM TCEP, 50% Glycerol, (pH 7.5 @

25°)

Specification Version: PS-M0654S v1.0

EnGen® Sau Cas9 Component List				
<b>NEB Part Number</b>	Component Description	Lot Number	Individual QC Result	
M0654SVIAL	EnGen® Sau Cas9	10040077	Pass	
B7203SVIAL	NEBuffer™ 3.1	10033149	Pass	

Assay Name/Specification	Lot # 10040102
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
Exonuclease Activity (Radioactivity Release) A 50 µl reaction in NEBuffer 3.1 containing 1 µg of a mixture of single and double-stranded [ ³H] 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
Functional Testing (Targeted Digestion) 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.	Pass
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.	Pass



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Assay Name/Specification	Lot # 10040102	
Protein Purity Assay (SDS-PAGE) EnGen® Sau Cas9 is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.	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	

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

Jessica Cane Production Scientist

20 Mar 2019

Michael Tonello

Packaging Quality Control Inspector

01 Apr 2019

