

New England Biolabs Certificate of Analysis

Product Name: I-SceI
Catalog Number: R0694L
Concentration: 5,000 U/ml
Unit Definition: One unit is defined as the amount of enzyme required to cleave 1 µg of pGPS2 NotI-linearized Control Plasmid in rCutSmart Buffer in 1 hour at 37°C in a total reaction volume of 50 µl.
Packaging Lot Number: 10236435
Expiration Date: 02/2026
Storage Temperature: -80°C
Storage Conditions: 10 mM Tris-HCl, 300 mM NaCl, 1 mM DTT, 0.1 mM EDTA, 50% Glycerol, 500 µg/ml rAlbumin (pH 7.4 @ 25°C)
Specification Version: PS-R0694S/L v2.0

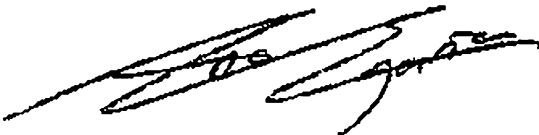
I-SceI Component List			
NEB Part Number	Component Description	Lot Number	Individual QC Result
R0694LVIAL	I-SceI	10228162	Pass
N0420SVIAL	pGPS2 NotI-linearized Control Plasmid	10232856	Pass
B6004SVIAL	rCutSmart™ Buffer	10229455	Pass

Assay Name/Specification	Lot # 10236435
Endonuclease Activity (Nicking) A 50 µl reaction in rCutSmart™ Buffer containing 1 µg of supercoiled PhiX174 DNA and a minimum of 15 units of I-SceI incubated for 4 hours at 37°C results in <20% conversion to the nicked form as determined by agarose gel electrophoresis.	Pass
Exonuclease Activity (Radioactivity Release) A 50 µl reaction in rCutSmart™ Buffer containing 1 µg of a mixture of single and double-stranded [³ H] E. coli DNA and a minimum of 50 units of I-SceI incubated for 4 hours at 37°C releases <0.1% of the total radioactivity.	Pass
Ligation and Recutting (Terminal Integrity) After a 10-fold over-digestion of pGPS2-NotI DNA with I-SceI, >95% of the DNA fragments can be ligated with T4 DNA ligase in 16 hours at 16°C. Of these ligated fragments, >95% can be recut with I-SceI.	Pass
Non-Specific DNase Activity (16 Hour) A 50 µl reaction in rCutSmart™ Buffer containing 1 µg of pGPS2-NotI DNA and a	Pass

Assay Name/Specification	Lot # 10236435
minimum of 50 units of I-SceI incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.	
Protein Purity Assay (SDS-PAGE) I-SceI is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.	Pass
qPCR DNA Contamination (E. coli Genomic) A minimum of 5 units of I-SceI is screened for the presence of E. coli genomic DNA using SYBR® Green qPCR with primers specific for the E. coli 16S rRNA locus. Results are quantified using a standard curve generated from purified E. coli genomic DNA. The measured level of E. coli genomic DNA contamination is ≤ 1 E. coli genome.	Pass

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

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Production Scientist
14 Mar 2024



Michael Tonello
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14 Mar 2024