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New England Biolabs Certificate of Analysis

Product Name: Pcil
Catalog Number: R0655L
Concentration: 10,000 U/ml

Unit Definition: One unit is defined as the amount of enzyme required to digest 1 µg

of pXba DNA in NEBuffer r3.1 in 1 hour at 37°C in a total reaction

volume of 50 μl.

Packaging Lot Number: 10236228
Expiration Date: 02/2026
Storage Temperature: -20°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-R0655S/L v2.0

Pcil Component List				
NEB Part Number	Component Description	Lot Number	Individual QC Result	
R0655LVIAL	Pcil	10228167	Pass	
B7024AVIAL	Gel Loading Dye, Purple (6X)	10221469	Pass	
B6003SVIAL	NEBuffer™ r3.1	10227733	Pass	

Assay Name/Specification	Lot # 10236228
Endonuclease Activity (Nicking) A 50 µl reaction in NEBuffer™ r3.1 containing 1 µg of supercoiled PhiX174 DNA and a minimum of 10 units of Pcil 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 NEBuffer™ r3.1 containing 1 μg of a mixture of single and double-stranded [³H] E. coli DNA and a minimum of 100 units of Pcil 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 pXba DNA with Pcil, >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 Pcil.	Pass
Non-Specific DNase Activity (16 Hour) A 50 μl reaction in NEBuffer™ r3.1 containing 1 μg of pXba DNA and a minimum of 50	Pass



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Assay Name/Specification	Lot # 10236228
units of Pcil 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) Pcil is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.	Pass
qPCR DNA Contamination (E. coli Genomic) A minimum of 10 units of Pcil 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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Ana Egana Production Scientist 18 Mar 2024 Michael Tonello

Packaging Quality Control Inspector

18 Mar 2024