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New England Biolabs Certificate of Analysis

Product Name: Xhol
Catalog Number: R0146L
Concentration: 20,000 U/ml

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

of Lambda DNA (HindIII digest) fragments in rCutSmart Buffer in 1

hour at 37°C in a total reaction volume of 50 μl.

Packaging Lot Number: 10162016
Expiration Date: 08/2024
Storage Temperature: -20°C

Storage Conditions: 10 mM Tris-HCl, 50 mM KCl, 1 mM DTT, 0.1 mM EDTA, 50% Glycerol, 200

μg/ml rAlbumin (pH 7.4 @ 25°C)

Specification Version: PS-R0146S/L/E v3.0

Xhol Component List				
NEB Part Number	Component Description	Lot Number	Individual QC Result	
R0146LVIAL	Xhol	10162009	Pass	
B7024AVIAL	Gel Loading Dye, Purple (6X)	10158559	Pass	
B6004SVIAL	rCutSmart™ Buffer	10158558	Pass	

Assay Name/Specification	Lot # 10162016
Blue-White Screening (Terminal Integrity)	Pass
A sample of Litmus 28i vector linearized with a 10-fold excess of Xhol, religated	
and transformed into an E. coli strain expressing the LacZ beta fragment gene	
results in <1% white colonies.	
Ligation and Recutting (Terminal Integrity)	Pass
After a 10-fold over-digestion of pXba DNA with Xhol, >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 XhoI.	
Protein Purity Assay (SDS-PAGE)	Pass
Xhol is >95% pure as determined by SDS PAGE analysis using Coomassie Blue detection.	
qPCR DNA Contamination (E. coli Genomic)	Pass
A minimum of 20 units of Xhol 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.	



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Assay Name/Specification	Lot # 10162016
The measured level of E. coli genomic DNA contamination is ≤ 1 E. coli genome.	
Non-Specific DNase Activity (16 Hour) A 50 µl reaction in rCutSmart™ Buffer containing 1 µg of Lambda-HindIII DNA and a minimum of 100 units of Xhol incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation 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 [³H] E. coli DNA and a minimum of 100 units of Xhol incubated for 4 hours at 37°C releases <0.1% of the total radioactivity.	Pass
Functional Testing (15 minute Digest) A 50 μl reaction in rCutSmart™ Buffer containing 1 μg of Lambda-HindIII DNA and 1 μl of Xhol incubated for 15 minutes at 37°C results in complete digestion as determined by agarose gel electrophoresis.	Pass
Endonuclease Activity (Nicking) A 50 µl reaction in rCutSmart™ Buffer containing 1 µg of supercoiled pBR322 DNA and a minimum of 100 units of Xhol incubated for 4 hours at 37°C results in <10% conversion to the nicked form as determined by agarose gel electrophoresis.	Pass

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

One or more products referenced in this document may be covered by a 3rd-party trademark. Please visit www.neb.com/trademarks for additional information.

YunJie Sun \
Production Scientist
30 Aug 2022

Michael Tonello

Packaging Quality Control Inspector

11 Oct 2022

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