

240 County Road Ipswich, MA 01938-2723 Tel 978-927-5054 Fax 978-921-1350 www.neb.com info@neb.com

## New England Biolabs Certificate of Analysis

Product Name: AfIIII

Catalog Number: R0541S

Concentration: 10,000 U/ml

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

of Lambda DNA in NEBuffer™ r3.1 in 1 hour at 37°C in a total

reaction volume of 50 μl.

Packaging Lot Number: 10137185
Expiration Date: 01/2024
Storage Temperature: -20°C

Storage Conditions: 10 mM Tris-HCl, 500 mM NaCl ,1 mM DTT , 0.1 mM EDTA , 50 % Glycerol

, 200 μg/ml rAlbumin (pH 7.4 @ 25°C)

Specification Version: PS-R0541S/L v2.0

AfIIII Component List				
<b>NEB Part Number</b>	Component Description	Lot Number	Individual QC Result	
R0541SVIAL	AfIIII	10137114	Pass	
B6003SVIAL	NEBuffer™ r3.1	10126635	Pass	

Assay Name/Specification	Lot # 10137185
Protein Purity Assay (SDS-PAGE)	Pass
AfIIII is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.	
qPCR DNA Contamination (E. coli Genomic)	Pass
A minimum of 10 units of AfIIII 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.	
Non-Specific DNase Activity (16 Hour)	Pass
A 50 μl reaction in NEBuffer™ r3.1 containing 1 μg of Lambda DNA and a minimum of	
30 units of AfIIII incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.	
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Functional Testing (15 minute Digest)	Pass
A 50 μl reaction in NEBuffer <sup>™</sup> r3.1 containing 1 μg of Lambda DNA and 1 μl of AfIIII	
incubated for 15 minutes at 37°C results in complete digestion as determined by	



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Assay Name/Specification	Lot # 10137185
agarose gel electrophoresis.	
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 50 units of AfIIII incubated for 4 hours at 37°C releases <0.1% of the total radioactivity.	Pass
Ligation and Recutting (Terminal Integrity)  After a 5-fold over-digestion of Lambda DNA with AfIIII, >95% of the DNA fragments can be ligated with T4 DNA ligase in 16 hours at 16°C. Of these ligated fragments,	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.

Penghaa Zhang Production Scientist

04 Mar 2022

Mary Negal

Packaging Quality Control Inspector

04 Mar 2022



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