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## New England Biolabs Certificate of Analysis

Product Name: HpyCH4III
Catalog Number: R0618L
Concentration: 5,000 U/ml

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

of Lambda DNA in 1 hour at 37°C in a total reaction volume of 50 μl.

Packaging Lot Number: 10168515
Expiration Date: 10/2024
Storage Temperature: -20°C

Storage Conditions: 100 mM NaCl, 10 mM Tris-HCl (pH 7.4), 1 mM DTT, 0.1 mM EDTA, 50%

Glycerol, 200 µg/ml BSA

Specification Version: PS-R0618S/L v2.0

HpyCH4III Component List				
<b>NEB Part Number</b>	Component Description	Lot Number	Individual QC Result	
R0618LVIAL	HpyCH4III	10168516	Pass	
B6004SVIAL	rCutSmart™ Buffer	10162783	Pass	

Assay Name/Specification	Lot # 10168515
DNase Activity (Labeled Oligo, 3' extension) A 50 µl reaction in CutSmart® Buffer containing a 20 nM solution of a fluorescent labeled double-stranded oligonucleotide containing a 3' extension and a minimum of 25 units of HpyCH4III incubated for 16 hours at 37°C yields <5% degradation as determined by capillary electrophoresis.	Pass
Double Stranded DNase Activity (Labeled Oligo) A 50 µl reaction in CutSmart® Buffer containing a 20 nM solution of a fluorescent labeled double-stranded oligonucleotide containing a blunt end and a minimum of 25 units of HpyCH4III incubated for 16 hours at 37°C yields <5% degradation as determined by capillary electrophoresis.	Pass
DNase Activity (Labeled Oligo, 5' extension) A 50 µl reaction in CutSmart® Buffer containing a 20 nM solution of a fluorescent labeled double-stranded oligonucleotide containing a 5' extension and a minimum of 25 units of HpyCH4III incubated for 16 hours at 37°C yields <5% degradation as determined by capillary electrophoresis.	Pass
Non-Specific DNase Activity (16 Hour)	Pass



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Assay Name/Specification	Lot # 10168515
A 50 µl reaction in CutSmart™ Buffer containing 1 µg of Lambda DNA and a minimum of 5 units of HpyCH4III incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.	
Ligation and Recutting (Terminal Integrity)  After a 5-fold over-digestion of Lambda DNA with HpyCH4III, ~50% 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 HpyCH4III.	Pass
Single Stranded DNase Activity (FAM-Labeled Oligo)  A 50 µl reaction in CutSmart® Buffer containing a 20 nM solution of a fluorescent internal labeled oligonucleotide and a minimum of 25 units of HpyCH4III incubated for 16 hours at 37°C yields <5% degradation as determined by capillary electrophoresis.	Pass

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

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YunJie Sun \
Production Scientist

19 Oct 2022

Michael Tonello

Packaging Quality Control Inspector

19 Oct 2022



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