

## New England Biolabs Certificate of Analysis

**Product Name:** Nsil-HF®  
**Catalog Number:** R3127S  
**Concentration:** 20,000 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:** 10233738  
**Expiration Date:** 02/2026  
**Storage Temperature:** -20°C  
**Storage Conditions:** 300 mM NaCl, 10 mM Tris-HCl (pH 7.4), 1 mM DTT, 0.1 mM EDTA, 50 % Glycerol, 500 µg/ml BSA  
**Specification Version:** PS-R3127S/L v1.0

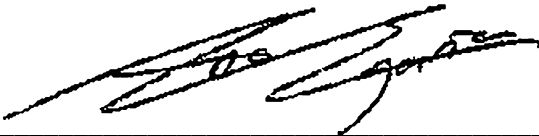
Nsil-HF® Component List			
NEB Part Number	Component Description	Lot Number	Individual QC Result
R3127SVIAL	Nsil-HF®	10206729	Pass
B7024AVIAL	Gel Loading Dye, Purple (6X)	10221469	Pass
B6004SVIAL	rCutSmart™ Buffer	10228580	Pass

Assay Name/Specification	Lot # 10233738
<b>Endonuclease Activity (Nicking)</b> A 50 µl reaction in CutSmart™ Buffer containing 1 µg of supercoiled φX174 DNA and a minimum of 100 units of Nsil-HF incubated for 4 hours at 37°C results in <10% conversion to the nicked form as determined by agarose gel electrophoresis.	Pass
<b>Exonuclease Activity (Radioactivity Release)</b> A 50 µl reaction in CutSmart™ Buffer containing 1 µg of a mixture of single and double-stranded [ <sup>3</sup> H] E. coli DNA and a minimum of 100 units of Nsil-HF incubated for 4 hours at 37°C releases <0.1% of the total radioactivity.	Pass
<b>Functional Test (15 minute Digest)</b> A 50 µl reaction in CutSmart™ Buffer containing 1 µg of Lambda DNA and 1 µl of Nsil-HF incubated for 15 minutes at 37°C results in complete digestion as determined by agarose gel electrophoresis.	Pass
<b>Ligation and Recutting (Terminal Integrity)</b> After a 20-fold over-digestion of Lambda DNA with Nsil-HF, >95% of the DNA fragments can be ligated with T4 DNA ligase in 16 hours at 16°C. Of these ligated fragments,	Pass

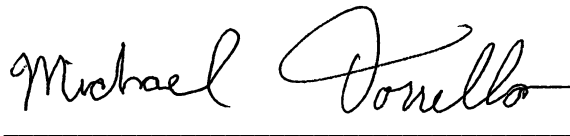
Assay Name/Specification	Lot # 10233738
>95% can be recut with Nsil-HF.	
<p><b>Non-Specific DNase Activity (16 Hour)</b> A 50 µl reaction in CutSmart™ Buffer containing 1 µg of Lambda DNA and a minimum of 100 units of Nsil-HF incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.</p>	<b>Pass</b>
<p><b>Protein Purity Assay (SDS-PAGE)</b> Nsil-HF is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.</p>	<b>Pass</b>

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](http://www.neb.com/trademarks) for additional information.



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22 Feb 2024



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22 Feb 2024