

New England Biolabs Certificate of Analysis

Product Name: *SpeI*
Catalog Number: *R0133M*
Concentration: *50,000 U/ml*
Unit Definition: *One unit is defined as the amount of enzyme required to digest 1 µg of pXba-XbaI DNA in 1 hour at 37°C in a total reaction volume of 50 µl.*
Packaging Lot Number: *10115908*
Expiration Date: *07/2023*
Storage Temperature: *-20°C*
Storage Conditions: *250 mM NaCl, 10 mM Tris-HCl (pH 7.4), 1 mM DTT, 0.1 mM EDTA, 50% Glycerol, 0.15% Triton® X-100, 200 µg/ml BSA*
Specification Version: *PS-R0133T/M v2.0*

SpeI Component List			
NEB Part Number	Component Description	Lot Number	Individual QC Result
R0133M VIAL	SpeI	10115907	Pass
B7024A VIAL	Gel Loading Dye, Purple (6X)	10093123	Pass
B6004S VIAL	rCutSmart™ Buffer	10121395	Pass

Assay Name/Specification	Lot # 10115908
Exonuclease Activity (Radioactivity Release) A 50 µl reaction in CutSmart® Buffer containing 1 µg of a mixture of single and double-stranded [³ H] E. coli DNA and a minimum of 50 units of SpeI incubated for 4 hours at 37°C releases <0.1% of the total radioactivity.	Pass
Non-Specific DNase Activity (16 Hour) A 50 µl reaction in CutSmart® Buffer containing 1 µg of pXba-XbaI digested DNA and a minimum of 50 units of SpeI incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.	Pass
Protein Purity Assay (SDS-PAGE) SpeI is >95% pure as determined by SDS PAGE analysis using Coomassie Blue detection.	Pass
Ligation and Recutting (Terminal Integrity) After a 20-fold over-digestion of T7 DNA with SpeI, >95% of the DNA fragments can be ligated with T4 DNA ligase in 16 hours at 16°C. Of these ligated fragments, >95% can	Pass

Assay Name/Specification	Lot # 10115908
<p>be recut with SpeI.</p> <p>Blue-White Screening (Terminal Integrity) A sample of LITMUS28 vector linearized with a 10-fold excess of SpeI, religated and transformed into an E. coli strain expressing the LacZ beta fragment gene results in <1% white colonies.</p>	<p>Pass</p>
<p>Endonuclease Activity (Nicking) A 50 µl reaction in CutSmart[®] Buffer containing 1 µg of supercoiled PhiX174 DNA and a minimum of 50 units of SpeI incubated for 4 hours at 37°C results in <20% conversion to the nicked form as determined by agarose gel electrophoresis.</p>	<p>Pass</p>

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.



Penghua Zhang
Production Scientist
14 Oct 2021



Josh Hersey
Packaging Quality Control Inspector
14 Oct 2021