

## New England Biolabs Certificate of Analysis

**Product Name:** XmnI  
**Catalog Number:** R0194S  
**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:** 10119488  
**Expiration Date:** 09/2023  
**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-R0194S/L/V v2.0

XmnI Component List			
NEB Part Number	Component Description	Lot Number	Individual QC Result
R0194SVIAL	XmnI	10119487	Pass
B7024AVIAL	Gel Loading Dye, Purple (6X)	10119053	Pass
B6004SVIAL	rCutSmart™ Buffer	10119384	Pass

Assay Name/Specification	Lot # 10119488
<b>Blue-White Screening (Terminal Integrity)</b> A sample of pUC19 vector linearized with a 10-fold excess of XmnI, religated and transformed into an E. coli strain expressing the LacZ beta fragment gene results in <1% white colonies.	Pass
<b>Ligation and Recutting (Terminal Integrity)</b> After a 20-fold over-digestion of Lambda DNA with XmnI, ~75% 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 XmnI.	Pass
<b>Protein Purity Assay (SDS-PAGE)</b> XmnI is >95% pure as determined by SDS PAGE analysis using Coomassie Blue detection.	Pass
<b>Non-Specific DNase Activity (16 Hour)</b> A 50 µl reaction in rCutSmart™ Buffer containing 1 µg of Lambda DNA and a minimum of 100 units of XmnI incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.	Pass

Assay Name/Specification	Lot # 10119488
<p><b>Functional Testing (15 minute Digest)</b> A 50 µl reaction in rCutSmart™ Buffer containing 1 µg of Lambda DNA and 1 µl of XmnI incubated for 15 minutes at 37°C results in complete digestion as determined by agarose gel electrophoresis.</p>	Pass
<p><b>Exonuclease Activity (Radioactivity Release)</b> A 50 µl reaction in rCutSmart™ 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 XmnI incubated for 4 hours at 37°C releases &lt;0.1% of the total radioactivity.</p>	Pass
<p><b>Endonuclease Activity (Nicking)</b> A 50 µl reaction in rCutSmart™ Buffer containing 1 µg of supercoiled LITMUS38i DNA and a minimum of 60 units of XmnI incubated for 4 hours at 37°C results in &lt;10% conversion to the nicked form as determined by agarose gel electrophoresis.</p>	Pass
<p><b>Endonuclease Activity (Nicking)</b> A 50 µl reaction in rCutSmart™ Buffer containing 1 µg of supercoiled LITMUS38i DNA and a minimum of 60 units of XmnI incubated for 4 hours at 37°C results in &lt;10% conversion to the nicked form as determined by agarose gel electrophoresis.</p>	Pass
<p><b>Exonuclease Activity (Radioactivity Release)</b> A 50 µl reaction in rCutSmart™ 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 XmnI incubated for 4 hours at 37°C releases &lt;0.1% of the total radioactivity.</p>	Pass
<p><b>Functional Testing (15 minute Digest)</b> A 50 µl reaction in rCutSmart™ Buffer containing 1 µg of Lambda DNA and 1 µl of XmnI incubated for 15 minutes at 37°C results in complete digestion as determined by agarose gel electrophoresis.</p>	Pass
<p><b>Non-Specific DNase Activity (16 Hour)</b> A 50 µl reaction in rCutSmart™ Buffer containing 1 µg of Lambda DNA and a minimum of 100 units of XmnI incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.</p>	Pass
<p><b>Protein Purity Assay (SDS-PAGE)</b> XmnI is &gt;95% pure as determined by SDS PAGE analysis using Coomassie Blue detection.</p>	Pass
<p><b>Ligation and Recutting (Terminal Integrity)</b> After a 20-fold over-digestion of Lambda DNA with XmnI, ~75% of the DNA fragments can be ligated with T4 DNA ligase in 16 hours at 16°C. Of these ligated fragments, &gt;95% can be recut with XmnI.</p>	Pass

Assay Name/Specification	Lot # 10119488
<p><b>Blue-White Screening (Terminal Integrity)</b> A sample of pUC19 vector linearized with a 10-fold excess of XmnI, religated and transformed into an E. coli strain expressing the LacZ beta fragment gene results in &lt;1% white colonies.</p>	<p><b>Pass</b></p>

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

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05 Oct 2021




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05 Oct 2021