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## New England Biolabs Product Specification

Product Name:	XbaI
Catalog #:	R0145T/M
Concentration:	100,000 units/ml
Unit Definition:	One unit is defined as the amount of enzyme required to digest 1 $\mu$ g of Lambda DNA (dam-/HindIII digest) in 1 hour at 37°C in a total reaction volume of 50 $\mu$ l.
Shelf Life:	24 months
Storage Temp:	-20°C
Storage Conditions:	10 mM Tris-HCl, 50 mM NaCl, 1 mM DTT, 0.1 mM EDTA, 50% Glycerol, 200 μg/ml BSA, (pH 7.4 @ 25°C)
Specification Version:	<i>PS-R0145T/M v2.0</i>
Effective Date:	12 Jul 2021

Assay Name/Specification (minimum release criteria)

**Blue-White Screening (Terminal Integrity)** - A sample of pUC19 vector linearized with a 10-fold excess of XbaI, religated and transformed into an *E. coli* strain expressing the LacZ beta fragment gene results in <1% white colonies.

Endonuclease Activity (Nicking) - A 50  $\mu$ l reaction in CutSmart® Buffer containing 1  $\mu$ g of supercoiled PhiX174 DNA and a minimum of 100 units of XbaI incubated for 4 hours at 37°C results in <10% conversion to the nicked form as determined by agarose gel electrophoresis.

**Exonuclease Activity (Radioactivity Release)** - A 50  $\mu$ l reaction in CutSmart® Buffer containing 1  $\mu$ g of a mixture of single and double-stranded [<sup>3</sup>H] *E. coli* DNA and a minimum of 200 units of XbaI incubated for 4 hours at 37°C releases <0.1% of the total radioactivity.

**Ligation and Recutting (Terminal Integrity)** - After a 10-fold over-digestion of pBC4XS DNA with XbaI, >95% 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 XbaI.

Non-Specific DNase Activity (16 Hour) - A 50 µl reaction in CutSmart® Buffer containing 1 µg of Lambda-HindIII dam- DNA and a minimum of 200 units of XbaI incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.

Protein Purity Assay (SDS-PAGE) - XbaI is >95% pure as determined by SDS PAGE analysis using Coomassie Blue detection.

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Date 12 Jul 2021

Derek Robinson Director, Quality Control



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