

240 County Road Ipswich, MA 01938-2723 Tel 978-927-5054 Fax 978-921-1350 www.neb.com info@neb.com

## New England Biolabs Product Specification

Product Name:	KasI
Catalog #:	R0544S/L
Concentration:	5,000 units/ml
Unit Definition:	One unit is defined as the amount of enzyme required to digest 1 $\mu$ g of pBR322 DNA in 1 hour at 37°C in a total reaction volume of 50 $\mu$ l.
Shelf Life:	12 months
Storage Temp:	-80°C
Storage Conditions:	500 mM KCl, 20 mM Tris-HCl (pH 7.0), 0.1 mM EDTA, 1mM MgCl <sub>2</sub> , 50% Glycerol, 0.10% Triton X-100, 200 μg/ml BSA
Specification Version:	<i>PS-R0544S/L v3.0</i>
Effective Date:	02 Nov 2021

## Assay Name/Specification (minimum release criteria)

**Blue-White Screening (Terminal Integrity)** - A sample of LITMUS38i vector linearized with a 10-fold excess of KasI, religated and transformed into an *E. coli* strain expressing the LacZ beta fragment gene results in <1% white colonies.

**Exonuclease Activity (Radioactivity Release)** - A 50  $\mu$ l reaction in CutSmart<sup>TM</sup> Buffer containing 1  $\mu$ g of a mixture of single and double-stranded [<sup>3</sup>H] *E. coli* DNA and a minimum of 5 units of KasI incubated for 4 hours at 37°C releases <0.1% of the total radioactivity.

**Ligation and Recutting (Terminal Integrity)** - After a 20-fold over-digestion of pBR322 DNA with KasI, >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 KasI.

Non-Specific DNase Activity (16 Hour) - A 50  $\mu$ l reaction in CutSmart<sup>TM</sup> Buffer containing 1  $\mu$ g of pBR322 DNA and a minimum of 5 Units of KasI 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) - KasI is >95% pure as determined by SDS PAGE analysis using Coomassie Blue detection.

\* The BSA in this product has been granted an EDQM "Certificate of Suitability" from the European Directorate for the Quality of Medicines (# R1-CEP-2003-204-Rev00) and has been granted a USDA Certificate for Export of Bovine Blood Plasma/Serum for Manufacture into Pharmaceutical Products.

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Date 02 Nov 2021

Derek Robinson Director, Quality Control



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