

## New England Biolabs Certificate of Analysis

**Product Name:** *KasI*  
**Catalog Number:** *R0544L*  
**Concentration:** *5,000 U/ml*  
**Unit Definition:** *One unit is defined as the amount of enzyme required to digest 1 µg of pBR322 DNA in rCutSmart Buffer in 1 hour at 37°C in a total reaction volume of 50 µl.*  
**Packaging Lot Number:** *10239973*  
**Expiration Date:** *04/2025*  
**Storage Temperature:** *-80°C*  
**Storage Conditions:** *10 mM Tris-HCl, 100 mM KCl, 1 mM DTT, 0.1 mM EDTA, 200 µg/ml rAlbumin, 50% Glycerol, (pH 7.4 @ 25°C)*  
**Specification Version:** *PS-R0544S/L v4.0*

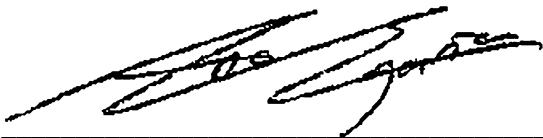
KasI Component List			
NEB Part Number	Component Description	Lot Number	Individual QC Result
R0544LVIAL	KasI	10239972	Pass
B6004SVIAL	rCutSmart™ Buffer	10235561	Pass

Assay Name/Specification	Lot # 10239973
<p><b>Blue-White Screening (Terminal Integrity)</b>            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 &lt;1% white colonies.</p>	<b>Pass</b>
<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 5 units of KasI incubated for 4 hours at 37°C releases &lt;0.1% of the total radioactivity.</p>	<b>Pass</b>
<p><b>Ligation and Recutting (Terminal Integrity)</b>            After a 20-fold over-digestion of pBR322 DNA with KasI, &gt;95% 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 KasI.</p>	<b>Pass</b>
<p><b>Non-Specific DNase Activity (16 Hour)</b>            A 50 µl reaction in rCutSmart™ Buffer containing 1 µg 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</p>	<b>Pass</b>

Assay Name/Specification	Lot # 10239973
detectable nuclease degradation as determined by agarose gel electrophoresis.	
<b>Protein Purity Assay (SDS-PAGE)</b> KasI is >95% pure as determined by SDS PAGE analysis using Coomassie Blue detection.	<b>Pass</b>
<b>qPCR DNA Contamination (E. coli Genomic)</b> A minimum of 5 units of KasI is screened for the presence of E. coli genomic DNA using SYBR® Green qPCR with primers specific for the E. coli 16S rRNA locus. Results are quantified using a standard curve generated from purified E. coli genomic DNA. The measured level of E. coli genomic DNA contamination is $\leq 1$ E. coli genome.	<b>Pass</b>

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

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21 May 2024



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