

## New England Biolabs Certificate of Analysis

**Product Name:** *Thermostable FEN1*  
**Catalog Number:** *M0645S*  
**Concentration:** *32,000 U/ml*  
**Unit Definition:** *One unit is defined as the amount of FEN1 required to cleave 10 pmol of 5' DNA flap containing oligonucleotide substrate in a total reaction volume of 10 µl for 10 minutes at 65°C.*  
**Packaging Lot Number:** *10197834*  
**Expiration Date:** *07/2025*  
**Storage Temperature:** *-20°C*  
**Storage Conditions:** *10 mM Tris-HCl , 100 mM KCl , 1 mM DTT , 0.1 mM EDTA , 0.1 % Triton®X-100 , 50 % Glycerol, (pH 7.4 @ 25°C)*  
**Specification Version:** *PS-M0645S/L v1.0*


Thermostable FEN1 Component List			
NEB Part Number	Component Description	Lot Number	Individual QC Result
M0645SVIAL	Thermostable FEN1	10197833	Pass
B9004SVIAL	ThermoPol® Reaction Buffer Pack	10187437	Pass

Assay Name/Specification	Lot # 10197834
<p><b>Endonuclease Activity (Nicking)</b>            A 50 µl reaction in ThermoPol® Reaction Buffer containing 1 µg of supercoiled PhiX174 DNA and a minimum of 160 units of Thermostable FEN1 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 ThermoPol® Reaction Buffer containing 1 µg of a mixture of single and double-stranded [<sup>3</sup>H] E. coli DNA and a minimum of 160 units of Thermostable FEN1 incubated for 4 hours at 37°C releases &lt;0.1% of the total radioactivity.</p>	Pass
<p><b>Non-Specific DNase Activity (16 Hour)</b>            A 50 µl reaction in ThermoPol® Reaction Buffer containing 1 µg of Lambda-HindIII DNA and a minimum of 320 units of Thermostable FEN1 incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by gel electrophoresis.</p>	Pass

Assay Name/Specification	Lot # 10197834
<p><b>Protein Purity Assay (SDS-PAGE)</b> Thermostable FEN1 is <math>\geq 95\%</math> pure as determined by SDS-PAGE analysis using Coomassie Blue detection.</p>	<b>Pass</b>
<p><b>RNase Activity (Extended Digestion)</b> A 10 <math>\mu</math>l reaction in NEBuffer 4 containing 40 ng of a 300 base single-stranded RNA and a minimum of 1 <math>\mu</math>l of Thermostable FEN1 is incubated at 37°C. After incubation for 16 hours, &gt;90% of the substrate RNA remains intact as determined by gel electrophoresis using fluorescent detection.</p>	<b>Pass</b>

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

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06 Jul 2023