

## New England Biolabs Product Specification

<i>Product Name:</i>	<i>Thermostable FEN1</i>
<i>Catalog #:</i>	<i>M0645S/L</i>
<i>Concentration:</i>	<i>32,000 units/ml</i>
<i>Unit Definition:</i>	<i>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.</i>
<i>Shelf Life:</i>	<i>24 months</i>
<i>Storage Temp:</i>	<i>-20°C</i>
<i>Storage Conditions:</i>	<i>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)</i>
<i>Specification Version:</i>	<i>PS-M0645S/L v1.0</i>
<i>Effective Date:</i>	<i>17 Nov 2017</i>

### Assay Name/Specification (minimum release criteria)

**Endonuclease Activity (Nicking)** - 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 <10% conversion to the nicked form as determined by agarose gel electrophoresis.

**Exonuclease Activity (Radioactivity Release)** - 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 <0.1% of the total radioactivity.

**Non-Specific DNase Activity (16 Hour)** - 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.

**Protein Purity Assay (SDS-PAGE)** - Thermostable FEN1 is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.

**RNase Activity (Extended Digestion)** - A 10 µl reaction in NEBuffer 4 containing 40 ng of a 300 base single-stranded RNA and a minimum of 1 µl of Thermostable FEN1 is incubated at 37°C. After incubation for 16 hours, >90% of the substrate RNA remains intact as determined by gel electrophoresis using fluorescent detection.



Date 17 Nov 2017

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Director of Quality Control

