

## New England Biolabs Product Specification

<i>Product Name:</i>	<i>DNA Gyrase (E. coli)</i>
<i>Catalog #:</i>	<i>M0306S/L</i>
<i>Concentration:</i>	<i>5,000 units/ml</i>
<i>Unit Definition:</i>	<i>One unit is defined as the amount of enzyme that catalyzes the conversion of 0.5 µg DNA Gyrase (E. coli) Substrate to &gt;95% supercoiled plasmid in a total reaction volume of 30 µl in 30 minutes at 37°C.</i>
<i>Shelf Life:</i>	<i>12 months</i>
<i>Storage Temp:</i>	<i>-80°C</i>
<i>Storage Conditions:</i>	<i>10 mM Tris-HCl, 50 mM KCl, 2 mM DTT, 0.1 mM EDTA, 50 % Glycerol, (pH 7.5 @ 25°C)</i>
<i>Specification Version:</i>	<i>PS-M0306S/L v1.0</i>
<i>Effective Date:</i>	<i>17 Jun 2016</i>

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

**Endonuclease Activity (Nicking)** - A 50 µl reaction in CutSmart® Buffer containing 1 µg of supercoiled pUC19 DNA and a minimum of 25 units of DNA Gyrase (*E. coli*) 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 CutSmart® Buffer containing 1 µg of a mixture of single and double-stranded [<sup>3</sup>H] *E. coli* DNA and a minimum of 25 units of DNA Gyrase (*E. coli*) 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 CutSmart® Buffer containing 1 µg of Lambda-HindIII DNA and a minimum of 25 units of DNA Gyrase (*E. coli*) incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.

**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 DNA Gyrase (*E. coli*) 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 Jun 2016

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Director of Quality Control

