

## New England Biolabs Product Specification

<i>Product Name:</i>	<i>CpG Methylated HeLa Genomic DNA</i>
<i>Catalog #:</i>	<i>N4007S</i>
<i>Concentration:</i>	<i>100 µg/ml</i>
<i>Unit Definition:</i>	<i>N/A</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, 1 mM EDTA, (pH 7.5 @ 25°C)</i>
<i>Specification Version:</i>	<i>PS-N4007S v1.0</i>
<i>Effective Date:</i>	<i>11 Apr 2016</i>

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

**A260/A280 Assay** - The ratio of UV absorption of CpG Methylated HeLa Genomic DNA at 260 and 280 nm is between 1.8 and 2.0.

**DNA Concentration (A260)** - The concentration of CpG Methylated HeLa Genomic DNA is between 100 and 110 µg/ml as determined by UV absorption at 260 nm.

**Electrophoretic Pattern (Genomic DNA)** - The banding pattern of CpG Methylated HeLa Genomic DNA on a 1.2% agarose gel is evaluated against a control lot for relative integrity and intensity as determined by gel electrophoresis using Ethidium Bromide.

**Functional Test (Methylation Specific PCR)** - CpG Methylated HeLa Genomic DNA was bisulfite converted, amplified by PCR with primers specific for methylated versus unmethylated PTEN or Rb promoter DNA, resulting in the expected product only with the methylated-specific primer sets.

**Functional Testing (Genomic DNA Methylation, Radioactivity Incorporation)** - A 50 µl reaction in NEBuffer 2, including [ <sup>3</sup>H ] AdoMet, containing 2.5 µg CpG Methylated HeLa Genomic DNA and 8 units of CpG Methylase (M.SssI) incubated for 4 hours at 37°C results in incorporation of <0.1% of the total radioactivity.

**Non-Specific DNase Activity (Genomic DNA, 16 hour)** - A 50 µl reaction in 1X NEBuffer 2 containing 2.5 µg of CpG Methylated HeLa Genomic DNA incubated for 16 hours at 37°C does not produce any further detectable nuclease degradation as determined by agarose gel electrophoresis.

**Restriction Digest (CpG Resistant)** - A 50 µl reaction in 1X CutSmart® Buffer containing 2.5 µg of CpG Methylated HeLa Genomic DNA and a minimum of 10 units of HpaII incubated for 1 hour at 37°C results in no detectable digestion of the DNA as determined by agarose gel electrophoresis.



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<b>Restriction Digest (CpG Sensitive)</b> - A 50 µl reaction in 1X CutSmart <sup>®</sup> Buffer containing 2.5 µg of CpG Methylated HeLa Genomic DNA and a minimum of 20 units of MspI incubated for 1 hour at 37°C produces the expected fragmentation pattern as determined by agarose gel electrophoresis.
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Date 11 Apr 2016

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Director of Quality Control

