

## New England Biolabs Product Specification

<i>Product Name:</i>	<i>NEBNext<sup>®</sup> Enzymatic 5hmC-seq Kit</i>
<i>Catalog #:</i>	<i>E3350G</i>
<i>Kit Components:</i>	<i>Control DNA Unmethylated Lambda (E7123) — Store at -20°C</i> <i>Control DNA 5hmC T4 (E3349) — Store at -20°C</i> <i>NEBNext<sup>®</sup> Ultra<sup>™</sup> II End Prep Reaction Buffer (E7647) — Store at -20°C</i> <i>NEBNext<sup>®</sup> Ultra<sup>™</sup> II End Prep Enzyme Mix (E7646) — Store at -20°C</i> <i>NEBNext<sup>®</sup> Ultra<sup>™</sup> II Ligation Master Mix (E7648) — Store at -20°C</i> <i>NEBNext<sup>®</sup> Ligation Enhancer (E7374) — Store at -20°C</i> <i>NEBNext<sup>®</sup> E5hmC-seq<sup>™</sup> Adaptor (E3366) — Store at -20°C</i> <i>NEBNext<sup>®</sup> Carrier DNA (E3351) — Store at -20°C</i> <i>Elution Buffer (E7124) — Store at -20°C</i> <i>NEBNext<sup>®</sup> Glucosylation Reaction Buffer (E3352) — Store at -20°C</i> <i>UDP-Glucose (E3353) — Store at -20°C</i> <i>T4-BGT (E3354) — Store at -20°C</i> <i>Stop Reagent (E7132) — Store at -20°C</i> <i>Deamination Reaction Buffer (E3356) — Store at -20°C</i> <i>Recombinant Albumin (E3357) — Store at -20°C</i> <i>APOBEC (E7133) — Store at -20°C</i> <i>NEBNext<sup>®</sup> Q5U<sup>®</sup> Master Mix (E3369) — Store at -20°C</i> <i>NEBNext<sup>®</sup> Sample Purification Beads (E3355) — Store at 25°C</i>
<i>Shelf Life:</i>	<i>12 months</i>
<i>Storage Temp:</i>	<i>Multi-temperature</i>
<i>Specification Version:</i>	<i>PS-E3350G v1.0</i>
<i>Effective Date:</i>	<i>17 Jan 2024</i>

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

**Functional Testing (E5hmC-seq<sup>™</sup> Library Construction)** - Each set of reagents is functionally validated and compared to the previous lot through construction of libraries made from genomic DNA and DNA controls (5hmC T4 and unmethylated Lambda), that are required for assessment of 5hmC. The kit's minimum and maximum DNA input requirements are used to make libraries that are sequenced on the same Illumina<sup>®</sup> flow cell. Library assessment is based on metrics including library yields, GC bias, insert size, and the percent 5hmC detected for CpG, CHG, CHH contexts within the genomic DNA and internal controls.





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\* **Individual Product Component Note** - Standard Quality Control Tests are performed for each component included in NEBNext® Enzymatic 5hmC-seq Kit and meet the designated specifications.

*One or more products referenced in this document may be covered by a 3rd-party trademark.  
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Date 17 Jan 2024

Lauren Brown  
Quality Approver

