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## New England Biolabs Product Specification

Product Name: Diluent A (with rAlbumin)

Catalog #: B8532S

Concentration: 1X Concentrate
Shelf Life: 36 months
Storage Temp: -20°C

Composition (1X): 10 mM Tris-HCl, 50 mM KCl, 1 mM DTT, 0.1 mM EDTA, 200 µg/ml rAlbumin, 50 % Glycerol, (pH 7.4 @)

25°C)

Specification Version: PS-B8532S v1.0 Effective Date: 11 Aug 2023

## Assay Name/Specification (minimum release criteria)

Endonuclease Activity (Nicking) - A 50  $\mu$ l reaction in rCutSmart<sup>TM</sup> Buffer containing 1  $\mu$ g of supercoiled PhiX174 DNA and a minimum of 10  $\mu$ l of Diluent A (with rAlbumin) incubated for 4 hours at 37°C results in <10% conversion to the nicked form as determined by agarose gel electrophoresis.

Non-Specific DNase Activity (16 Hour) - A 50  $\mu$ l reaction in rCutSmart<sup>TM</sup> Buffer containing 1  $\mu$ g of PhiX174-HaeIII DNA and a minimum of 10  $\mu$ l of Diluent A (with rAlbumin) incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.

pH (buffers/solutions) - The pH of 1X Diluent A (with rAlbumin) is between pH 7.3 and 7.5 at 25°C.

qPCR DNA Contamination (*E. coli* Genomic) - A minimum of 1  $\mu$ l of Diluent A (with rAlbumin) is screened for the presence of *E. coli* genomic DNA using SYBR® Green qPCR with primers specific for the *E. coli* 16S rRNA locus. Results are quantified using a standard curve generated from purified *E. coli* genomic DNA. The measured level of *E. coli* genomic DNA contamination is  $\leq 1$  *E. coli* genome.

RNase Activity (Extended Digestion) - A 10  $\mu$ l reaction in NEBuffer 4 containing 40 ng of a 300 base single-stranded RNA and a minimum of 1  $\mu$ l of Diluent A (with rAlbumin) 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.

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Lauren Brown Quality Approver







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Date 11 Aug 2023