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Date

29 Sep 2016

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New England Biolabs Product Specification

Product Name: Isothermal Amplification Buffer Pack

Catalog #: B0537S

Concentration: 10X Concentrate

Shelf Life: 36 months
Storage Temp: -20°C

Composition (1X): 20 mM Tris-HCl, 50 mM KCl, 10 mM (NH₄)₂SO₄, 2 mM MgSO₄, 0.1 % Tween® 20, (pH 8.8 @ 25°C)

Specification Version: PS-B0537S v1.0

Effective Date: 29 Sep 2016

Assay Name/Specification (minimum release criteria)

Endonuclease Activity (Nicking, Buffer) - A 50 μ l reaction in 2X Isothermal Amplification Buffer containing 1 μ g of supercoiled PhiX174 DNA 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, Buffer) - A 50 μ l reaction in 2X Isothermal Amplification Buffer containing 1 μ g of T3 DNA in addition to a reaction containing Lambda-HindIII DNA 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 10X Isothermal Amplification Buffer is between pH 8.7 and 8.9 at 25°C.

Phosphatase Activity (pNPP, Buffer) - A 200 μ l reaction in 1M Diethanolamine @ pH 9.8 and 0.5 mM MgCl₂ containing 2.5 mM *p*-Nitrophenyl Phosphate (pNPP) and a minimum of 40 μ l Isothermal Amplification Buffer incubated for 4 hours at 37°C yields <0.0001 unit of alkaline phosphatase activity as determined by spectrophotometric analysis.

qPCR DNA Contamination (*E. coli* Genomic, Buffer) - A minimum of 1 μ l of Isothermal Amplification Buffer 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 ≤ 1 *E. coli* genome.

RNAse Activity Assay (4 Hour Digestion) - A 10 μ l reaction in NEBuffer 4 containing 40 ng of a 300 base single-stranded RNA and a minimum of 1 μ l of Isothermal Amplification Buffer is incubated at 37°C. After incubation for 4 hours, >90% of the substrate RNA remains intact as determined by gel electrophoresis using fluorescent detection.

Derek Robinson

Director of Quality Control





