240 County Road Ipswich, MA 01938-2723 Tel 978-927-5054 Fax 978-921-1350 www.neb.com info@neb.com

## New England Biolabs Certificate of Analysis

Product Name: Bsu DNA Polymerase, Large Fragment

Catalog #: M0330S/L
Concentration: 5,000 units/ml

Unit Definition: One unit is defined as the amount of enzyme that will incorporate 10 nmol of dNTP into acid insoluble material in 30 minutes

at 37°C.

 Lot #:
 0041603

 Assay Date:
 03/2016

 Expiration Date:
 03/2018

 Storage Temp:
 -20°C

Storage Conditions: 25 mM Tris-HCl, 50 mM NaCl, 1 mM DTT, 0.1 mM EDTA, 50 % Glycerol, (pH 7.4 @ 25°C)

Specification Version: PS-M0330S/L v1.0

Effective Date: 16 Oct 2015

Assay Name/Specification (minimum release criteria)	Lot #0041603
<b>Endonuclease Activity (Nicking)</b> - A 50 μl reaction in NEBuffer 2 containing 1 μg of supercoiled PhiX174 DNA and a minimum of 50 units of <i>Bsu</i> DNA Polymerase, Large Fragment incubated for 4 hours at 37°C results in <10% conversion to the nicked form as determined by agarose gel electrophoresis.	Pass
<b>Exonuclease Activity (Radioactivity Release)</b> - A 50 $\mu$ l reaction in NEBuffer 2 containing 1 $\mu$ g of a mixture of single and double-stranded [ $^3$ H] <i>E. coli</i> DNA and a minimum of 50 units of <i>Bsu</i> DNA Polymerase, Large Fragment incubated for 4 hours at 37°C releases <0.1% of the total radioactivity.	Pass
Non-Specific DNase Activity (16 Hour) - A 50 µl reaction in NEBuffer 2 containing 1 µg of T3 DNA in addition to a reaction containing Lambda-HindIII DNA and a minimum of 5 units of <i>Bsu</i> DNA Polymerase, Large Fragment incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.	Pass
<b>Phosphatase Activity (pNPP)</b> - A 200 μl reaction in 1M Diethanolamine, pH 9.8, 0.5 mM MgCl <sub>2</sub> containing 2.5 mM <i>p</i> -Nitrophenyl Phosphate (pNPP) and a minimum of 100 units <i>Bsu</i> DNA Polymerase, Large Fragment incubated for 4 hours at 37°C yields <0.0001 unit of alkaline phosphatase activity as determined by spectrophotometric analysis.	Pass
<b>Protein Purity Assay (SDS-PAGE)</b> - <i>Bsu</i> DNA Polymerase, Large Fragment is ≥ 97% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.	Pass









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Assay Name/Specification (minimum release criteria)	Lot #0041603
<b>qPCR DNA Contamination (</b> <i>E. coli</i> <b>Genomic)</b> - A minimum of 5 units of $Bsu$ DNA Polymerase, Large Fragment is screened for the presence of <i>E. coli</i> genomic DNA using SYBR® Green qPCR with primers specific for the <i>E. coli</i> 16S rRNA locus. Results are quantified using a standard curve generated from purified <i>E. coli</i> genomic DNA. The measured level of <i>E. coli</i> genomic DNA contamination is $\leq 1$ <i>E. coli</i> genome.	Pass
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 <i>Bsu</i> DNA Polymerase, Large Fragment 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.	Pass
Single Stranded DNase Activity (FAM-Labeled Oligo) - A 50 μl reaction in NEBuffer 2 containing a 10 nM solution of a fluorescent internal labeled oligonucleotide and a minimum of 50 units of <i>Bsu</i> DNA Polymerase, Large Fragment incubated for 30 minutes at 37°C yields <10% degradation as determined by capillary electrophoresis.	Pass

Authorized by Melanie Fortier 16 Oct 2015







Inspected by
Lea Antonopoulos
02 Jun 2016