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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:	Isothermal Amplification Buffer II Pack
Catalog Number:	B0374S
Concentration:	10 X Concentrate
Lot Number:	10036128
Expiration Date:	12/2021
Storage Temperature:	-20°C
Specification Version:	PS-B0374S v2.0
Composition (1X):	20 mM Tris-HCl, 10 mM (NH4)2SO4, 150 mM KCl, 2 mM MgSO4, 0.1 % Tween® 20, (pH 8.8 @ 25°C)

Isothermal Amplification Buffer II Pack Component List				
NEB Part Number	Component Description	Lot Number	Individual QC Result	
B0374SVIAL	Isothermal Amplification Buffer II Pack	10029501	Pass	

Assay Name/Specification	Lot # 10036128
<b>Endonuclease Activity (Nicking, Buffer)</b> A 50 µl reaction in 2X Isothermal Amplification Buffer II containing 1 µg 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.	Pass
<b>Non-Specific DNase Activity (16 hour, Buffer)</b> A 50 µl reaction in 2X Isothermal Amplification Buffer II containing 1 µg 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.	Pass
<b>pH (buffers/solutions)</b> The pH of 10X Isothermal Amplification Buffer II is between pH 8.7 and 8.9 at 25°C.	Pass
<b>Phosphatase Activity (pNPP, Buffer)</b> A 200 µl 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 µl 10X Isothermal Amplification Buffer II incubated for 4 hours at 37°C yields <0.0001 unit of alkaline phosphatase activity as determined by spectrophotometric analysis.	Pass
<b>qPCR DNA Contamination (E. coli Genomic, Buffer)</b> A minimum of 1 μl of Isothermal Amplification Buffer II is screened for the presence	Pass





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Assay Name/Specification	Lot # 10036128
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.	
<b>RNAse Activity Assay (4 Hour Digestion)</b> 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 Isothermal Amplification Buffer II incubated for 4 hours at 37°C results in no detectable degradation of the RNA as determined by gel electrophoresis using fluorescent detection.	Pass

This product has been tested and shown to be in compliance with all specifications.

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Tony Spear-Alfonso Production Scientist 07 Jan 2019

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Michael Tonello Packaging Quality Control Inspector 13 Feb 2019

