

## New England Biolabs Certificate of Analysis

**Product Name:** NEBuffer™ 4  
**Catalog #:** B7004S  
**Concentration:** 10X Concentrate  
**Lot #:** 0131611  
**Assay Date:** 11/2016  
**Expiration Date:** 11/2019  
**Storage Temp:** -20°C  
**Composition (1X):** 50 mM Potassium Acetate, 20 mM Tris-acetate, 10 mM Magnesium Acetate, 1 mM DTT, (pH 7.9 @ 25°C)  
**Specification Version:** PS-B7004S v1.0  
**Effective Date:** 27 Mar 2018

Assay Name/Specification (minimum release criteria)	Lot #0131611
<b>Conductivity (buffers/solutions)</b> - The conductivity of 10X NEBuffer 4 is between 36 and 54 mS at 25°C.	<b>Pass</b>
<b>Endonuclease Activity (Nicking, Buffer)</b> - A 50 µl reaction in 1X NEBuffer 4 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.	<b>Pass</b>
<b>Functional Testing (Restriction Digest, BSA, Buffer)</b> - A 50 µl reaction in 1X NEBuffer 4 plus 100 µg/ml Bovine Serum Albumin containing 1 µg of Lambda DNA and 1 unit of MscI incubated for 1 hour at 37°C results in complete digestion of the substrate DNA as determined by agarose gel electrophoresis.	<b>Pass</b>
<b>Functional Testing (Restriction Digest, BSA, Buffer)</b> - A 50 µl reaction in 1X NEBuffer 4 plus 100 µg/ml Bovine Serum Albumin containing 1 µg of Lambda dam- DNA and 1 unit of ClaI incubated for 1 hour at 37°C results in complete digestion of the substrate DNA as determined by agarose gel electrophoresis.	<b>Pass</b>
<b>Non-Specific DNase Activity (16 hour, Buffer)</b> - A 50 µl reaction in 1X NEBuffer 4 containing 1 µg of PhiX174-HaeIII DNA incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.	<b>Pass</b>
<b>pH (buffers/solutions)</b> - The pH of 10X NEBuffer 4 is between pH 7.8 and 8.0 at 25°C.	<b>Pass</b>
<b>RNase Activity (Buffer)</b> - A 10 µl reaction in 1X NEBuffer 4 containing 40 ng of a 300 base single-stranded RNA is incubated at 37°C. After incubation for 16 hours, >90% of the substrate RNA remains intact as determined by fluorescent detection.	<b>Pass</b>



Authorized by  
Doreen Duquette  
27 Mar 2018



Inspected by  
Tony Spear-Alfonso  
14 Dec 2016

