

## New England Biolabs Certificate of Analysis

**Product Name:** Vent® DNA Polymerase  
**Catalog Number:** M0254S  
**Concentration:** 2,000 U/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 75°C.  
**Packaging Lot Number:** 10165003  
**Expiration Date:** 09/2024  
**Storage Temperature:** -20°C  
**Storage Conditions:** 10 mM Tris-HCl , 100 mM KCl , 1 mM DTT , 0.1 mM EDTA , 0.1 % Triton®X-100 , 50 % Glycerol, (pH 7.4 @ 25°C)  
**Specification Version:** PS-M0254S/L v1.0

Vent® DNA Polymerase Component List			
NEB Part Number	Component Description	Lot Number	Individual QC Result
M0254SVIAL	Vent® DNA Polymerase	10164135	Pass
B9004SVIAL	ThermoPol® Reaction Buffer Pack	10165338	Pass
B1003SVIAL	Magnesium Sulfate (MgSO <sub>4</sub> ) Solution	10159437	Pass

Assay Name/Specification	Lot # 10165003
<p><b>Endonuclease Activity (Nicking, Polymerase, dNTP)</b>            A 50 µl reaction in ThermoPol® Reaction Buffer in the presence of 400 µM dNTPs containing 1 µg of supercoiled pUC19 DNA and a minimum of 20 units of Vent® DNA Polymerase incubated for 4 hours at 37°C results in &lt;10% conversion to the nicked form as determined by agarose gel electrophoresis.</p>	Pass
<p><b>Protein Purity Assay (SDS-PAGE)</b>            Vent® DNA Polymerase is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.</p>	Pass
<p><b>qPCR DNA Contamination (E. coli Genomic)</b>            A minimum of 2 units of Vent® DNA Polymerase 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.</p>	Pass
<p><b>RNase Activity (Extended Digestion)</b></p>	Pass

Assay Name/Specification	Lot # 10165003
<p>A 10 µl reaction in NEBuffer 4 containing 40 ng of a 300 base single-stranded RNA and a minimum of 1 µl of Vent® DNA Polymerase is incubated at 37°C. After incubation for 16 hours, &gt;90% of the substrate RNA remains intact as determined by gel electrophoresis using fluorescent detection.</p>	
<p><b>PCR Amplification (2.0 kb Lambda DNA)</b> A 25 µl reaction in ThermoPol® Reaction Buffer in the presence of 200 µM dNTPs and 0.5 µM primers containing 5 ng Lambda DNA with 0.25 units of Vent® DNA Polymerase for 25 cycles of PCR amplification results in the expected 2.0 kb product.</p>	<b>Pass</b>
<p><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 p-Nitrophenyl Phosphate (pNPP) and a minimum of 100 units Vent® DNA Polymerase incubated for 4 hours at 37°C yields &lt;0.0001 unit of alkaline phosphatase activity as determined by spectrophotometric analysis.</p>	<b>Pass</b>

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

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Trinh Nguyen  
Production Scientist  
19 Sep 2022



Michael Tonello  
Packaging Quality Control Inspector  
12 Oct 2022