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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:	Vent® (exo-) DNA Polymerase
Catalog Number:	M0257S
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:	10172269
Expiration Date:	10/2024
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-M0257S/L v2.0

Vent® (exo-) DNA Polymerase Component List				
NEB Part Number	Component Description	Lot Number	Individual QC Result	
M0257SVIAL	Vent® (exo-) DNA Polymerase	10171034	Pass	
B9004SVIAL	ThermoPol® Reaction Buffer Pack	10165338	Pass	
B1003SVIAL	Magnesium Sulfate (MgSO ₄) Solution	10159437	Pass	

Assay Name/Specification	Lot # 10172269
PCR Amplification (2.0 kb Lambda DNA) A 25 μ I reaction in ThermoPol® Reaction Buffer in the presence of 200 μ M dNTPs and 0.2 μ M primers containing 5 ng Lambda DNA with 0.5 units of Vent® (exo-) DNA Polymerase for 30 cycles of PCR amplification results in the expected 2.0 kb product.	Pass
Phosphatase Activity (pNPP) A 200 µl reaction in 1M Diethanolamine, pH 9.8, 0.5 mM MgCl2 containing 2.5 mM p-Nitrophenyl Phosphate (pNPP) and a minimum of 100 units Vent® (exo-) DNA Polymerase incubated for 4 hours at 37°C yields <0.0001 unit of alkaline phosphatase activity as determined by spectrophotometric analysis.	Pass
qPCR DNA Contamination (E. coli Genomic) A minimum of 2 units of Vent® (exo-) 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.	Pass





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Assay Name/Specification	Lot # 10172269
RNase Activity (Extended Digestion) A 10 μ I reaction in NEBuffer 4 containing 40 ng of a 300 base single-stranded RNA and a minimum of 1 μ I of Vent® (exo-) DNA Polymerase 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
Protein Purity Assay (SDS-PAGE) Vent® (exo-) DNA Polymerase is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.	Pass
Exonuclease Activity (Radioactivity Release) A 50 µl reaction in ThermoPol® Reaction Buffer containing 1 µg of a mixture of single and double-stranded [³ H] E. coli DNA and a minimum of 200 units of Vent® (exo-) DNA Polymerase incubated for 4 hours at 37°C and 75°C releases <0.1% of the total radioactivity.	Pass
Single Stranded DNase Activity (FAM-Labeled Oligo) A 20 µl reaction in ThermoPol® Reaction Buffer containing a 10 nM solution of a fluorescent internal labeled oligonucleotide and a minimum of 20 units of Vent® (exo-) DNA Polymerase incubated for 30 minutes at 37°C and 75°C yields <10% degradation as determined by capillary electrophoresis.	Pass
Endonuclease Activity (Nicking) A 50 µl reaction in ThermoPol® Reaction Buffer containing 1 µg of supercoiled PhiX174 DNA and a minimum of 200 units of Vent® (exo-) DNA Polymerase incubated for 4 hours at 37°C and 75°C results in <10% conversion to the nicked form as determined by agarose gel electrophoresis.	Pass
Non-Specific DNase Activity (16 Hour) A 50 µl reaction in NEBuffer 2 containing 1 µg of T3 or T7 DNA in addition to a reaction containing Lambda-HindIII DNA and a minimum of 2 units of Vent® (exo-) DNA Polymerase incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.	Pass

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

One or more products referenced in this document may be covered by a 3rd-party trademark. Please visit www.neb.com/trademarks for additional information.





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