

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: Thermostable FEN1

Catalog Number: M0645S
Concentration: 32,000 U/ml

Unit Definition: One unit is defined as the amount of FEN1 required to cleave 10 pmol

of 5' DNA flap containing oligonucleotide substrate in a total

reaction volume of 10 μl for 10 minutes at 65°C.

Packaging Lot Number: 10222850
Expiration Date: 11/2025
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-M0645S/L v1.0

| Thermostable FEN1 Component List | | | | |
|----------------------------------|---------------------------------|------------|----------------------|--|
| NEB Part Number | Component Description | Lot Number | Individual QC Result | |
| M0645SVIAL | Thermostable FEN1 | 10208868 | Pass | |
| B9004SVIAL | ThermoPol® Reaction Buffer Pack | 10215580 | Pass | |

| Assay Name/Specification | Lot # 10222850 |
|--|----------------|
| Endonuclease Activity (Nicking) A 50 μl reaction in ThermoPol® Reaction Buffer containing 1 μg of supercoiled PhiX174 DNA and a minimum of 160 units of Thermostable FEN1 incubated for 4 hours at 37°C results in <10% conversion to the nicked form as determined by agarose gel electrophoresis. | Pass |
| Exonuclease Activity (Radioactivity Release) A 50 μl reaction in ThermoPol® Reaction Buffer containing 1 μg of a mixture of single and double-stranded [³H] E. coli DNA and a minimum of 160 units of Thermostable FEN1 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 ThermoPol® Reaction Buffer containing 1 µg of Lambda-HindIII DNA and a minimum of 320 units of Thermostable FEN1 incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by gel electrophoresis. | Pass |



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| Assay Name/Specification | Lot # 10222850 |
|--|----------------|
| Protein Purity Assay (SDS-PAGE) Thermostable FEN1 is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection. | Pass |
| RNase Activity (Extended Digestion) A 10 µl reaction in NEBuffer 4 containing 40 ng of a 300 base single-stranded RNA and a minimum of 1 µl of Thermostable FEN1 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 |

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.

Production Scientist

Michael Tonello

Packaging Quality Control Inspector

04 Jan 2024



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