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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.

Lot Number: 10050210
Expiration Date: 04/2021
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 | 10043262 | Pass | |
| B9004SVIAL | ThermoPol® Reaction Buffer Pack | 10041932 | Pass | |

| Assay Name/Specification | Lot # 10050210 |
|--|----------------|
| Protein Purity Assay (SDS-PAGE) | Pass |
| Thermostable FEN1 is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie | |
| Blue detection. | |
| RNase Activity (Extended Digestion) | Pass |
| 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. | |
| | |
| Endonuclease Activity (Nicking) | Pass |
| 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. | |
| | |
| Exonuclease Activity (Radioactivity Release) | Pass |
| A 50 μl reaction in ThermoPol® Reaction Buffer containing 1 μg of a mixture of | |



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| Assay Name/Specification | Lot # 10050210 |
|--|----------------|
| 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. | |
| 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 |

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

Production Scientist

18 Apr 2019

Michael Tonello

Packaging Quality Control Inspector

22 Jul 2019

