

New England Biolabs Certificate of Analysis

Product Name: Afu Uracil-DNA Glycosylase (UDG)
Catalog Number: M0279S
Concentration: 2,000 U/ml
Unit Definition: One unit is defined as the amount of enzyme that catalyzes the release of 60 pmol of uracil per minute from double-stranded, uracil-containing DNA. Activity is measured by release of [³H]-uracil in a 50 µl reaction containing 0.2 µg DNA (104-105 cpm/µg) in 30 minutes at 65°C.
Packaging Lot Number: 10068175
Expiration Date: 03/2022
Storage Temperature: -20°C
Storage Conditions: 10 mM Tris-HCl, 50 mM KCl, 1 mM DTT, 0.1 mM EDTA, 0.1 mg/ml BSA, 50% Glycerol, pH 7.4 @ 25°C
Specification Version: PS-M0279S/L v3.0

Afu Uracil-DNA Glycosylase (UDG) Component List			
NEB Part Number	Component Description	Lot Number	Individual QC Result
M0279SVIAL	Afu Uracil-DNA Glycosylase (UDG)	10068173	Pass
B9005SVIAL	ThermoPol® II (Mg-free) Reaction Buffer Pack	0021803	Pass

Assay Name/Specification	Lot # 10068175
<p>DNase Activity (Labeled Oligo, 5' extension) A 50 µl reaction in CutSmart™ Buffer containing a 20 nM solution of a fluorescent labeled double-stranded oligonucleotide containing a 5' extension and a minimum of 10 units of Afu Uracil-DNA Glycosylase (UDG) incubated for 16 hours at 37°C yields <5% degradation as determined by capillary electrophoresis.</p>	Pass
<p>Double Stranded DNase Activity (Labeled Oligo) A 50 µl reaction in CutSmart™ Buffer containing a 20 nM solution of a fluorescent labeled double-stranded oligonucleotide containing a blunt end and a minimum of 10 units of Afu Uracil-DNA Glycosylase (UDG) incubated for 16 hours at 37°C yields <5% degradation as determined by capillary electrophoresis.</p>	Pass
<p>Single Stranded DNase Activity (FAM-Labeled Oligo) A 50 µl reaction in CutSmart™ Buffer containing a 20 nM solution of a fluorescent internal labeled oligonucleotide and a minimum of 10 units of Afu Uracil-DNA Glycosylase (UDG) incubated for 16 hours at 37°C yields <5% degradation as</p>	Pass

Assay Name/Specification	Lot # 10068175
determined by capillary electrophoresis.	
<p>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 2 units of Afu Uracil-DNA Glycosylase (UDG) is incubated at 37°C. After incubation for 16 hours, >90% of the substrate RNA remains intact as determined by gel electrophoresis using polyacrylamide gel electrophoresis detection.</p>	Pass
<p>Protein Purity Assay (SDS-PAGE) Afu Uracil-DNA Glycosylase (UDG) is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.</p>	Pass
<p>Protein Concentration (A280/NanoDrop) The concentration of Afu Uracil-DNA Glycosylase (UDG) is 0.027 mg/ml +/- 10% as determined by UV absorption at 280 nm. Protein concentration is determined by the Pace method using the extinction coefficient of 16,110 and molecular weight of 22,720 daltons for Afu Uracil-DNA Glycosylase (UDG) (Pace, C.N. et al. (1995) Protein Sci., 4, 2411-2423).</p>	Pass
<p>Non-Specific DNase Activity (16 Hour) A 50 ul reaction in ThermoPol Reaction Buffer containing 1 ug of Lambda DNA and a minimum of 50 units of Afu Uracil-DNA Glycosylase (UDG) incubated for 16 hours at 65°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.</p>	Pass
<p>Endonuclease Activity (Nicking) A 50 ul reaction in ThermoPol Reaction Buffer containing 1 ug of supercoiled PhiX174 DNA and a minimum of 4 units of Afu Uracil-DNA Glycosylase (UDG) incubated for 4 hours at 65°C results in <20% conversion to the nicked form as determined by agarose gel electrophoresis.</p>	Pass
<p>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 50 units of Afu Uracil-DNA Glycosylase (UDG) incubated for 4 hours at 65°C releases <0.1% of the total radioactivity.</p>	Pass
<p>DNase Activity (Labeled Oligo, 3' extension) A 50 µl reaction in CutSmart™ Buffer containing a 20 nM solution of a fluorescent labeled double-stranded oligonucleotide containing a 3' extension and a minimum of 10 units of Afu Uracil-DNA Glycosylase (UDG) incubated for 16 hours at 37°C yields <5% degradation as determined by capillary electrophoresis.</p>	Pass

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

Lauren Higgins

Lauren Sears Higgins
Production Scientist
10 Mar 2020

Michael Tonello

Michael Tonello
Packaging Quality Control Inspector
31 Mar 2020