

New England Biolabs Certificate of Analysis

Product Name: WarmStart® Afu Uracil-DNA Glycosylase (UDG)
Catalog Number: M1282S
Concentration: 2,000 U/ml
Unit Definition: One unit is defined as the amount of enzyme required to release 60 pmol per minute of a fluorescently labeled 47-mer single-stranded DNA oligonucleotide containing a single uracil base in 30 minutes at 65°C in a total reaction volume of 50 µl in 1X Thermopol II Buffer.
Packaging Lot Number: 10219302
Expiration Date: 11/2025
Storage Temperature: -20°C
Storage Conditions: 10 mM Tris-HCl, 50 mM KCl, 1 mM DTT, 0.1 mM EDTA, 0.1 mg/ml rAlbumin, 50% Glycerol (pH 7.4 @ 25°C)
Specification Version: PS-M1282S v1.0

WarmStart® Afu Uracil-DNA Glycosylase (UDG) Component List			
NEB Part Number	Component Description	Lot Number	Individual QC Result
M1282SVIAL	WarmStart® Afu Uracil-DNA Glycosylase (UDG)	10217763	Pass
B9005SVIAL	ThermoPol® II (Mg-free) Reaction Buffer Pack	10139210	Pass

Assay Name/Specification	Lot # 10219302
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.	Pass
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.	Pass
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.	Pass

Assay Name/Specification	Lot # 10219302
<p>Endonuclease Activity (Nicking) A 50 µl reaction in ThermoPol® Reaction Buffer containing 1 µg 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 rCutSmart™ 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>Functional Testing (WarmStart Inhibition) A 50 µl reaction in ThermoPol® II Buffer containing a minimum of 2 units of WarmStart® Afu Uracil-DNA Glycosylase (UDG) incubated for 30 minutes at 25°C releases <3 pmol per minute (<5% activity) of a fluorescently labeled 47-mer single-stranded DNA oligonucleotide containing a single uracil base.</p>	Pass
<p>Non-Specific DNase Activity (16 Hour) A 50 µl reaction in ThermoPol® Reaction Buffer containing 1 µg of Lambda DNA and a minimum of 20 units of WarmStart® Afu Uracil-DNA Glycosylase (UDG) incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.</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>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 WarmStart® 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 fluorescent detection.</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 determined by capillary electrophoresis.</p>	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.



Jamie Souza
Production Scientist
17 Nov 2023



Josh Hersey
Packaging Quality Control Inspector
24 Nov 2023