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USER® Enzyme 37°

Thermolabile USER II Enzyme ([NEB #M5508](#)) and Thermostable USER III Enzyme ([NEB #M5509](#)) are available.

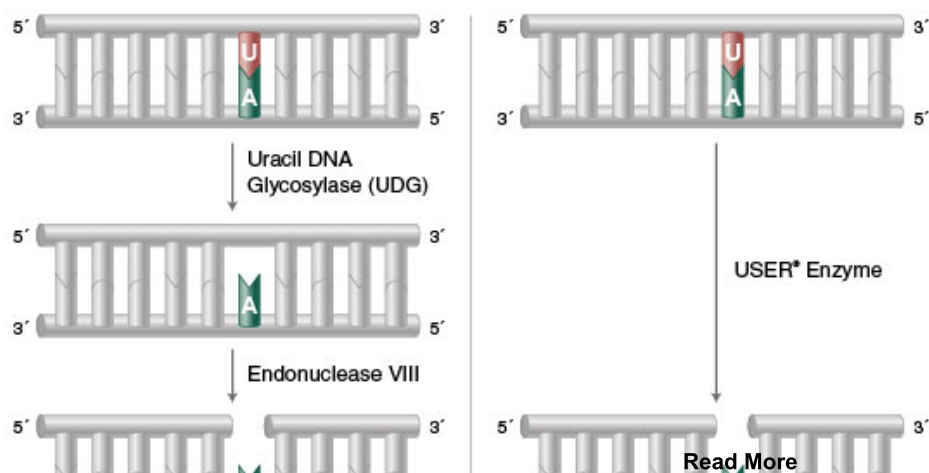
We are excited to announce that all reaction buffers are now BSA-free. NEB began switching our BSA-containing reaction buffers in April 2021 to buffers containing **Recombinant Albumin** (rAlbumin) for restriction enzymes and some DNA modifying enzymes. Find more details at www.neb.com/BSA-free.

USER (Uracil-Specific Excision Reagent) Enzyme (1) generates a single nucleotide gap at the location of a uracil residue.

Applications of USER include:

- Directional RNA Seq
- NEBNext adaptor cleavage
- USER cloning

Figure 1: USER Enzyme



Reagents Supplied

2,112 Product Citations

Featured Video



What are endonucleases and their applications?

Catalog #	Concentration	Size	List Price	Your Price	Quantity
M5505S	1,000 units/ml	50 units	\$83.00	Sign In	
M5505L	1,000 units/ml	250 units	\$337.00	Sign In	

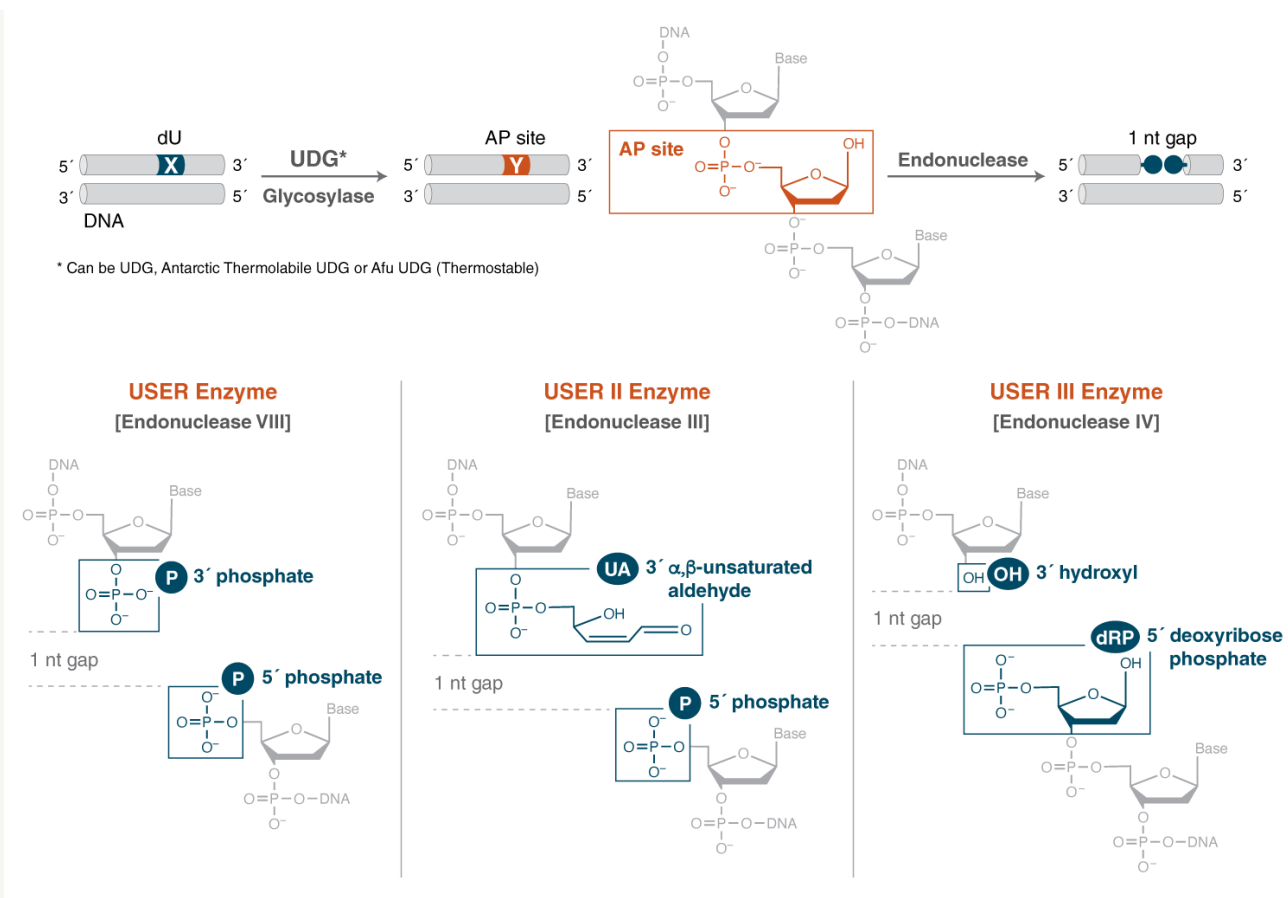
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Product Information

USER (Uracil-Specific Excision Reagent) Enzyme (1) generates a single nucleotide gap at the location of a uracil. USER Enzyme is a mixture of Uracil DNA glycosylase (UDG) and the DNA glycosylase-lyase Endonuclease VIII. UDG catalyses the excision of a uracil base, forming an abasic (apyrimidinic) site while leaving the phosphodiester backbone intact (2,3). The lyase activity of Endonuclease VIII breaks the phosphodiester backbone at the 3' and 5' sides of the abasic site so that base-free deoxyribose is released (4,5).

To help select the best DNA assembly method for your needs, please use our [Synthetic Biology/DNA Assembly Selection Chart](#).

Figure 2: USER Enzyme (NEB #M5505), Thermolabile USER II Enzyme (NEB #M5508) and Thermostable USER III Enzyme (NEB #M5509) generate different functional ends after cleavage of DNA.



In addition to different optimal reaction temperatures (37°C for USER and Thermolabile USER II Enzyme and 65°C for Thermostable USER III Enzyme) and ability to be heat inactivated (Thermolabile USER II Enzyme only), the different USER Enzymes generate different 3' and 5' termini after cleavage. USER Enzyme (NEB #M5505) contains Endonuclease VIII and leaves a 3' and 5' phosphate after cleavage. Thermolabile USER II Enzyme (NEB #M5508) contains Endonuclease III and leaves a 3'-phospho-α, β-unsaturated aldehyde and 5' phosphate after cleavage. Thermostable USER III Enzyme (NEB #M5509) contains Endonuclease IV and leaves a 3'-hydroxyl and 5'-deoxyribose phosphate.

Product Source

The two component proteins are purified separately from *E. coli* K-12 strains containing plasmids encoding Endonuclease VIII and Uracil-DNA Glycosylase.

This product is related to the following categories: DNA Repair Enzymes and Structure-specific Endonucleases Products

This product can be used in the following applications: Applications of USER® and Thermolabile USER II Enzymes, USER® Cloning, DNA Assembly and Cloning, | More +

Reagents Supplied

Reagents Supplied

The following reagents are supplied with this product:

NEB #	Component Name	Component #	Stored at (°C)	Amount	Concentration
M5505S			-20		
	USER® Enzyme	M5505SVIAL	-20	1 x 0.05 ml	1,000 units/ml
	rCutSmart™ Buffer	B6004SVIAL	-20	1 x 1.25 ml	10 X
M5505L			-20		
	USER® Enzyme	M5505LVIAL	-20	1 x 0.25 ml	1,000 units/ml
	rCutSmart™ Buffer	B6004SVIAL	-20	1 x 1.25 ml	10 X

Properties & Usage

Unit Definition

One unit is defined as the amount of enzyme required to nick 10 pmol of a 34 mer oligonucleotide duplex containing a single uracil base, in 15 minutes at 37°C in a total reaction volume of 10 µl.

Reaction Conditions

1X rCutSmart™ Buffer

Incubate at 37°C

1X rCutSmart™ Buffer

50 mM Potassium Acetate

20 mM Tris-acetate

10 mM Magnesium Acetate

100 µg/ml Recombinant Albumin

(pH 7.9 @ 25°C)

Storage Buffer

10 mM Tris-HCl

5 mM NaCl

1 mM DTT

0.1 mM EDTA

175 µg/ml BSA

50% Glycerol

50 mM KCl

pH 7.4 @ 25°C

Heat Inactivation

No

Related Products

Companion Products

- [Monarch® Plasmid Miniprep Kit](#)
- [Monarch® DNA Gel Extraction Kit](#)
- [Monarch® PCR & DNA Cleanup Kit \(5 µg\)](#)
- [Q5U® Hot Start High-Fidelity DNA Polymerase](#)

Product Notes

1. USER Enzyme is active in all commercial PCR buffers tested. It also has 100% activity in TE (10 mM Tris-HCl pH 8.0, 0.1 mM EDTA).
2. Incubate at 37°C.

References

1. Bitinaite, J. unpublished observations.
2. Lindhal, T., Ljungquist, S., Siegert, W., Nyberg, B. and Sperens, B. (1977). *J. Biol. Chem.* 252, 3286-3294.
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5. Jiang, D., Hatahet, Z., Melamede, R.J., Kow, Y.W. and Wallace, S.S. (1997). *J. Biol. Chem.* 272, 32230-32239.

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