

Product Information		
VeraSeq ULtra DNA Polymerase		
Part Number	P7520L	
Concentration	2,000 U/mL	
Unit Size	500 U	
Storage Temperature	-25ºC to -15ºC	
Lot Number		
Reference Number		

Product Specifications P7520L Rev 02

Product Description: VeraSeq ULtra DNA polymerase is an engineered, ultra-thermostable, uracil-literate polymerase designed to maximize the speed, accuracy, and length of DNA synthesis during sequencing template preparation. The result is a novel enzyme that can read through uracil, extend a kilobase of sequence in 15 seconds and has an accuracy 25 times higher than Taq DNA Polymerase.

Product Specifications				
P7520				
Assay	SDS Purity	Specific Activity	DS Endonuclease	E. coli DNA Contamination
Units Tested	n/a	n/a	120	150
Specification	>95%	100,000 U/mg	No Conversion	<10 copies

Source of Protein: A recombinant E. coli strain carrying the engineered VeraSeq Ultra gene.

<u>Unit Definition</u>: 1 unit is defined as the amount of enzyme required to incorporate 10 nmoles of dNTPs into acid-insoluble form at 74°C in 30 minutes.

Molecular weight: 97,754 Daltons

Quality Control Analysis:

Unit Activity is measured using a 2-fold serial dilution method. Dilutions of enzyme were made in 1X reaction buffer and added to 50 μL reactions containing activated calf thymus DNA; 25 mM TAPS (tris-[hydroxymethyl]-methyl-amino-propanesulfonic acid, sodium salt),pH 9.3 at 25°C; 50 mM KCl; 2 mM MgCl2; 1 mM ß-mercaptoethanol; 200 μM each dATP, dGTP, dTTP; and 100 μM [³H]-dCTP (0.075 Ci/mmole). Reaction vessels were mixed and incubated at 74°C for 10 minutes.

Protein Concentration (OD₂₈₀) is determined by OD₂₈₀ absorbance.

Physical Purity is evaluated by SDS-PAGE of concentrated and diluted enzyme solutions followed by silver stain detection. Purity is assessed by comparing the aggregate mass of contaminant bands in the concentrated sample to the mass of the protein of interest band in the diluted sample.

Double-Stranded Endonuclease is determined in a 50 μ L reaction containing 0.5 μ g of plasmid DNA and 10 μ L of enzyme solution incubated for 4 hours at 37°C.

E.coli 16S rDNA Contamination is evaluated using 5 µL replicate samples of enzyme solution denatured and screened in a TaqMan qPCR assay for the presence of contaminating *E.coli* genomic DNA using oligonucleotide primers corresponding to the 16S rRNA locus.

<u>Supplied in:</u> 20mM Tris-HCl, 100mM KCl, 1mM DTT, 0.1mM EDTA, Stabilizer, 50% glycerol pH 7.4 @ 25°C.
<u>Supplied with:</u>
5X VeraSeq Buffer II (B7102)
5X VeraSeq GC Buffer (B7130)



Extension Rate: 15 seconds per kilobase at 72°C

Fidelity: > 25X higher than Tag DNA Polymerase

Proofreading (3'-5' exo): Yes, strong Nick-translation (5'-3' exo): No

Thermostability: Highly thermostable Able to extend an RNA primer: No

Generate blunt end products: Yes

Kit Contents

Part Number	P7520L
Concentration	2,000 U/mL
Unit Size	500 U
5X VeraSeq buffer II	6 X 1.5 mL
5X VeraSeq GC buffer	3 X 1.5 mL

Common Applications

Ideal choice for applications requiring high fidelity DNA amplification, such as cloning, Next Generation sequencing, synthetic biology.

Protocol

General precautions should be taken when setting up a PCR, including setting up the reaction on ice, adding polymerase last, gentle pipetting, thorough mixing and a quick centrifugation. The following procedure can be used as a guideline. Reactions may need to be optimized individually.

VeraSeq 2.0 Ultra DNA Polymerase

Polymerase Properties

Strand Displacement: No

Extends from a nick: No

Uracil read through: Yes

Reaction setup (for 50 µL)*

Component	Volume (µL)	Final Concentration
Sterile H ₂ O	х	
5X VeraSeq buffer II or 5X VeraSeq GC buffer ¹	10	1X
10 mM dNTP mix ²	1	200 μM each
Primer 1 ³	х	0.2 μΜ
Primer 2 ³	х	0.2 μΜ
DNA template ⁴	х	See usage note #4
VeraSeq Ultra DNA Polymerase⁵	0.5	1 U

Typical Cycling Conditions**

Step	Temperature	Time	Cycles
Initial Denaturation	98°C	30 s	1
Denaturation	98°C	5-10 s	
Annealing	Varies	10-30 s	15-35
Extension	72°C	15-30 s/kb	
Final Extension	72°C	5-10 min	1
	4°C	hold	
** Cycling conditions may need to be optimized, depending on the			

* Total reaction volume can be adjusted as needed

Usage Notes:

1. 5X VeraSeq buffer II should be used as the default buffer for high-fidelity amplification. For GC-rich and difficult templates, use 5X VeraSeq GC buffer.

2. VeraSeq 2.0 ULtra DNA Polymerase can be used in PCR amplification to generate deoxy Uridine containing products by including dUTP in the reaction.

3. A final concentration of 0.2 μ M is recommended for each primer, but it can be varied in the range of 0.2 – 1 μ M.

4. Recommended template quantities:

Complexity	Source Example	Guideline
Low	Plasmid, Virus, BAC	1 pg – 10 ng
High	Genomic DNA	50 – 250 ng

5. One unit is usually sufficient for amplifying most targets. For long targets (>1 kb), difficult templates or to increase yield, it may be necessary to add up to 2 units of enzyme.

6. Both 5X VeraSeq buffer II and GC buffer are formulated to provide a final 1X concentration of MgCl2 of 1.5 mM. In cases where additional Mg2+ is required, adjust the final Mg2+ concentration in 0.2 mM steps.

7. For GC rich templates, DMSO may be used to reduce the secondary structure of complex templates. DMSO is generally used at a 3 % final concentration (v/v). If additional optimization is required, adjust the concentration in 1-2% increments (2-9% in final reaction). The primer annealing temperature should be lowered to account for the presence of the solvent.

8. VeraSeq ULtra DNA Polymerase is also compatible with other PCR-enhancing additives, such as BSA and betaine.

Limitations of Use

This product was developed, manufactured, and sold for *in vitro* use only. The product is not suitable for administration to humans or animals. SDS sheets relevant to this product are available upon request.