

Product Information	
<b>VeraSeq 2.0 High-Fidelity DNA Polymerase</b>	
Part Number	P7511L
Concentration	2,000 U/mL
Unit Size	500 U
Storage Temperature	-25°C to -15°C
Lot Number	
Reference Number	

**Product Description:** VeraSeq 2.0 High-Fidelity DNA polymerase is an engineered, ultra-thermostable polymerase designed to maximize the speed, accuracy, and length of DNA synthesis during sequencing template preparation. The result is a novel enzyme that can extend a kilobase of sequence in 15 seconds and with an accuracy 50 times higher than Taq DNA Polymerase.

Product Specifications				
P7511				
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 2.0 gene.

**Unit Definition:** 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:** 97,697 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 MgCl<sub>2</sub>; 1 mM β-mercaptoethanol; 200 µM each dATP, dGTP, dTTP; and 100 µM [<sup>3</sup>H]-dCTP (0.075 Ci/mmol). Reaction vessels were mixed and incubated at 74°C for 10 minutes.

**Protein Concentration (OD<sub>280</sub>)** is determined by OD<sub>280</sub> 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 µL reaction containing 0.5 µg of plasmid DNA and 10 µL 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.

**Supplied in:** 20mM Tris-HCl, 100mM KCl, 1mM DTT, 0.1mM EDTA, Stabilizer, 50% glycerol pH 7.4 @ 25°C.

**Supplied with:**

5X VeraSeq Buffer II (B7102)

5X VeraSeq GC Buffer (B7130)

**VeraSeq 2.0 High-Fidelity DNA Polymerase**

**Kit Contents**

Part Number	P7511L	P7511S
Concentration	2,000 U/mL	2,000 U/mL
Unit Size	500 U	100 U
5X VeraSeq buffer II	6 X 1.5 mL	1 X 1.5 mL
5X VeraSeq GC buffer	3 X 1.5 mL	1 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.

Reaction setup (for 50 µL)\*

Component	Volume (µL)	Final Concentration
Sterile H <sub>2</sub> O	x	
5X VeraSeq buffer II or 5X VeraSeq GC buffer <sup>1</sup>	10	1X
10 mM dNTP mix <sup>2</sup>	1	200 µM each
Primer 1 <sup>3</sup>	x	0.2 µM
Primer 2 <sup>3</sup>	x	0.2 µM
DNA template <sup>4</sup>	x	See usage note #4
VeraSeq 2.0 DNA Polymerase <sup>5</sup>	0.5	1 U

\* Total reaction volume can be adjusted as needed

**Usage Notes:**

- 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.
- VeraSeq 2.0 High-Fidelity DNA Polymerase stalls on uracil residues in the template strand and prevents further extension. Therefore, dUTP should not be used in the reaction. If DNA templates contains uracil or dUTP needs to be incorporated, use VeraSeq Ultra (P7520).
- A final concentration of 0.2 µM is recommended for each primer, but it can be varied in the range of 0.2 – 1 µM.
- Recommended template quantities:

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

- 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.
- Both 5X VeraSeq buffer II and GC buffer are formulated to provide a final 1X concentration of MgCl<sub>2</sub> of 1.5 mM. In cases where additional Mg<sup>2+</sup> is required, adjust the final Mg<sup>2+</sup> concentration in 0.2 mM steps.
- 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.
- VeraSeq 2.0 High-Fidelity DNA Polymerase is also compatible with other PCR-enhancing additives, such as BSA and betaine.

**Polymerase Properties**

Extension Rate: 15 seconds per kilobase at 72°C  
 Proofreading (3'-5' exo): Yes, strong  
 Nick-translation (5'-3' exo): No  
 Fidelity: > 50X higher than *Taq* DNA Polymerase  
 Strand Displacement: No  
 Thermostability: Highly thermostable  
 Able to extend an RNA primer: No  
 Extends from a nick: No  
 Generate blunt end products: Yes  
 Uracil read through: No

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 amplicon of interest

**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.