

# LONG AND ACCURATE

## Long and Accurate PCR Selection Guide

Catalog Number	Product Description	Proofreading Enzyme with 3'→5' Exonuclease Activity	Fidelity <sup>a</sup>	Amplification Length <sup>b</sup>	Direct Load <sup>c</sup>	Control Template and Primers	Assembled Master Mix <sup>d</sup>
D8045	AccuTaq LA DNA Polymerase	✓	up to 6.5×	0.1 to >20 (40) kb	—	—	—
D4812	REDAccuTaq LA DNA Polymerase	✓	up to 6.5×	0.1 to >20 (40) kb	✓	—	—
D5062	KlenTaq LA DNA Polymerase	✓	up to 4×	0.1 to >5 (10) kb	—	—	—
D1691	KlenTaq DV ReadyMix	✓	—	0.1 to >3 (1.5) kb	—	—	✓
D1816	RED KlenTaq DV ReadyMix	✓	—	0.1 to >3 (1.5) kb	✓	—	✓

a) Fidelity compared to Taq DNA Polymerase.

b) Two values are provided for the indicated amplification length. The range provided refers to the average length achieved from complex genomic targets, while the number in parentheses refers to lengths routinely achieved with less complicated targets such as plasmid or lambda phage DNA.

c) REDAccuTaq and RED KlenTaq contain an inert red dye. The dye provides visual confirmation that the polymerase has been added to the reaction and mixing is complete. Aliquots from the PCR can be directly loaded onto the gel without adding loading buffers or tracking dyes. The dye has no effect on automated DNA sequencing, ligation, transformation or other downstream applications.

d) Each ReadyMix is conveniently supplied at 2× and prepared using the indicated thermostable DNA polymerase, ultrapure 99%+ dNTPs, and high quality molecular biology reagents. To prepare a 50 µl PCR reaction, add 25 µl of the ReadyMix to 25 µl of water containing primers and template.

## AccuTaq™ LA DNA Polymerase

### Higher Fidelity for Specialized PCR Needs

AccuTaq LA DNA Polymerase is an optimized blend of Sigma's high quality Taq DNA Polymerase and a small amount of thermostable proofreading polymerase that exhibits 3'→5' exonuclease. By blending Taq with the right amount of this proofreading enzyme misincorporation errors are corrected, producing PCR amplicons that are longer and more accurate. This mix allows generation of amplicons from 0.25 kb to 40 kb.

### Features and Benefits

- Increased fidelity, up to 6.5× that of Taq DNA Polymerase
- Efficiently and accurately produce amplicons up to 22 kb on genomic templates and up to 40 kb on less complex templates such as lambda or bacterial genomic DNA

### Available in direct load

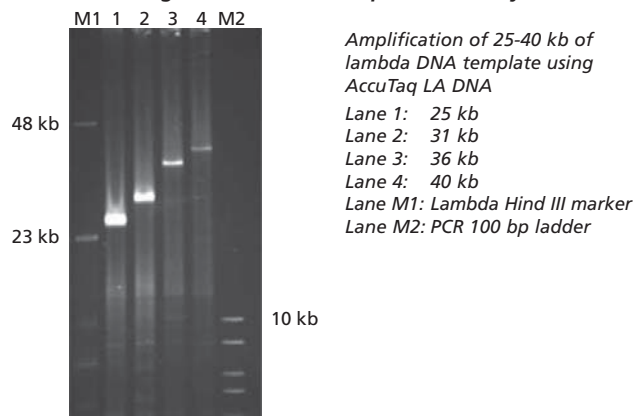
REDAccuTaq LA DNA Polymerase allows for quick recognition in high throughput applications as well as direct loading of amplification products onto agarose gels for electrophoresis. The inert red dye has no effect on automated sequencing, restriction enzyme digestion, ligation or other downstream manipulations. However, the PCR product is easily separated from the dye by standard purification methods.

**Components:** AccuTaq LA DNA Polymerase  
AccuTaq LA 10× Buffer  
Vial of dimethyl sulfoxide (DMSO) (D8045 only)

**Unit definition:** One unit incorporates 10 nmol of total dNTPs into acid-precipitable DNA in 30 min at 74 °C

**Storage:** -20 °C  
Shipped in wet ice

### Longer Products with Improved Fidelity



Cat. No.	Product Description	Quantity
D8045	AccuTaq LA DNA Polymerase 5 units per µl	125 units 500 units 1,500 units
D4812	REDAccuTaq LA DNA Polymerase 1 unit per µl	50 units 250 units

# LONG AND ACCURATE

## KlenTaq® LA DNA Polymerase Mix

### For Increased Yields and Fidelity

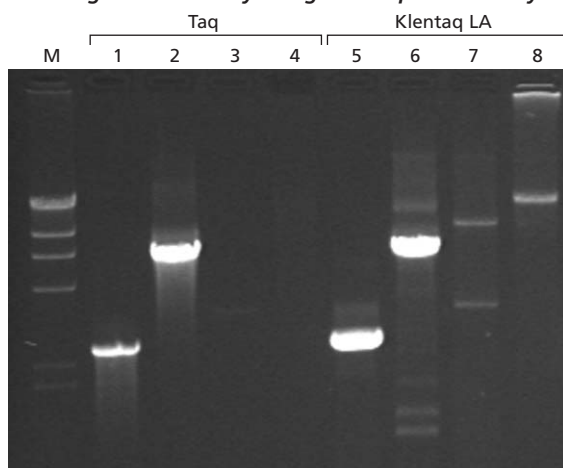
KlenTaq LA DNA Polymerase Mix is an optimized mixture combining KlenTaq-1 with a proofreading enzyme. KlenTaq-1 is a Klenow-fragment analog of Taq DNA Polymerase. It has no endonuclease or exonuclease activity, but is more thermostable than Taq or other terminal deletions of Taq. Since a wide range of magnesium concentration is tolerated by this enzyme, generally no magnesium optimization is needed. The proofreading polymerase provides the 3'→5' exonuclease activity that is necessary for longer and higher fidelity products.

KlenTaq LA DNA Polymerase provides an excellent alternative to Taq DNA Polymerase for intermediate length products. It allows higher yields and greater fidelity (up to 4× that of Taq). It can amplify genomic DNA up to 5 kb or up to 10 kb on less complex DNA targets such as bacterial, viral targets or cDNA. Its increased thermostability makes it the ideal choice for amplifying GC-rich regions or templates with difficult secondary structure.

### Features and Benefits

- KlenTaq LA DNA Polymerase has increased thermostability and processivity, resulting in increased yields
- Amplify difficult structure or GC-rich templates. The increased thermostability allows higher temperature conditions to disrupt difficult secondary structures
- Increased fidelity at up to 4× higher than that of Taq DNA Polymerase
- Tolerance to a broad range of magnesium concentrations eliminates the need to optimize  $MgCl_2$
- Amplify up to 5 kb genomic targets and up to 10 kb on less complex targets, such as lambda DNA

### Increase Length and Yield by Using KlenTaq LA DNA Polymerase



Lambda DNA was amplified using primer sets for 2.5 kb, 6 kb, 10 kb and 20 kb fragments. KlenTaq LA generates higher yields than Taq DNA Polymerase at 2.5 and 6 kb and is also able to amplify 10 and 20 kb fragments. Amount of lambda DNA template used for each PCR reaction: 1 ng for 2.5 kb, 2 ng for 6 kb, 2.5 ng for 10 kb, 50 ng for 20 kb.

Lane M: Marker  
Lanes 1, 5: 2.5 kb  
Lanes 2, 6: 6 kb  
Lanes 3, 7: 10 kb  
Lanes 4, 8: 20 kb

**Components:** KlenTaq LA DNA Polymerase  
KlenTaq LA 10× Buffer

**Unit definition:** One unit will incorporate 10 nmol of total dNTPs into acid-precipitable DNA in 30 min at 74 °C

**Concentration:** 5 units per  $\mu$ l

**Storage:** -20 °C  
Shipped in wet ice

### References

1. Barnes, W.M., PCR amplification of up to 35-kb DNA with high fidelity and high yield from bacteriophage templates. *Proc. Natl. Acad. Sci. USA* **91**, 2216-2220 (1994).

Cat. No.	Product Description	Quantity
D5062	KlenTaq LA DNA Polymerase Mix	125 units 500 units 1,500 units

# LONG AND ACCURATE

## KlenTaq® DV ReadyMix

KlenTaq DV ReadyMixes combine the performance benefits of KlenTaq DNA Polymerase with the convenience of a ReadyMix. This ready-to-use mixture of KlenTaq-1 DNA Polymerase (a 5'-exo-minus, N-terminal deletion of Taq DNA Polymerase), 99% pure deoxynucleotides, reaction buffer and a small amount of a proofreading DNA polymerase is provided in a 2× concentrate. RED KlenTaq DV ReadyMix includes Sigma's proprietary inert red dye.

For reaction set-up, add the ReadyMix (25 µl) to the primers, template and water (total volume 50 µl). Reaction volumes can be scaled down, if desired.

KlenTaq-1 is more efficient and more processive than either native Taq DNA Polymerase or other N-terminal deletions of Taq. This means greater yield is attained with the same number of cycles than when using Taq DNA Polymerase.

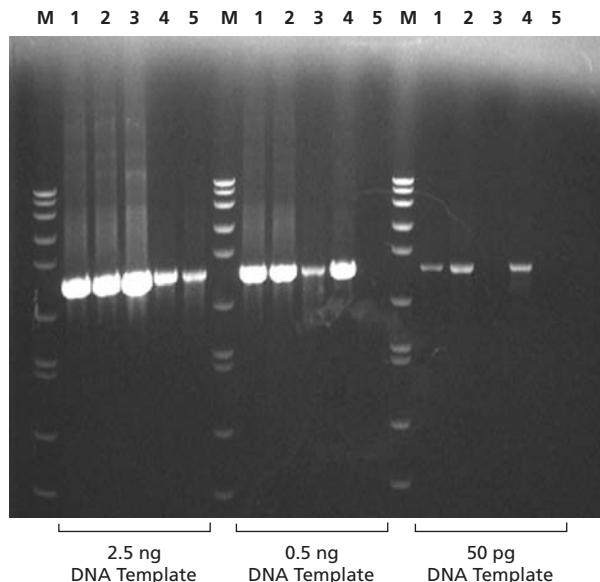
In RED KlenTaq DV ReadyMix, the inert red dye allows for easy confirmation of enzyme addition and visualization of complete mixing during setup. After the PCR reaction, the PCR product can be loaded directly onto an agarose gel. There is no need to add a loading buffer/tracking dye prior to electrophoresis. The red dye migrates at approximately the same rate as a 125 base pair fragment.

KlenTaq DV ReadyMix is an excellent choice for applications requiring high PCR yields from less complex, thoroughly purified templates.

### Features and Benefits

- Better efficiency and yield than Taq-based formulations, due to the higher processivity of KlenTaq-1
- Magnesium optimization is unnecessary, since KlenTaq works over a broad magnesium range
- Excellent amplification up to 3 kb for less complex templates (e.g. plasmid and lambda DNA) and 1.5 kb on human genomic DNA
- RED KlenTaq DV ReadyMix provides immediate confirmation that not only has the enzyme been added, but that proper mixing has occurred. An aliquot can be taken directly from the reaction and loaded onto an agarose gel for electrophoresis

### Greater Sensitivity and Yield with KlenTaq Blends



KlenTaq DV ReadyMixes and long and accurate enzymes from suppliers S and A were used to amplify a 2.5 kb fragment of lambda DNA.

Lane 1: RED KlenTaq DV ReadyMix

Lane 2: KlenTaq DV ReadyMix

Lane 3: Taq DNA Polymerase

Lane 4: Supplier S, enzyme Y

Lane 5: Supplier A, enzyme A

Lane M: Wide Range DNA marker

**Components:** KlenTaq DV and RED KlenTaq DV ReadyMixes provided in one convenient tube as a 2× concentrate

**Unit definition:** One unit will incorporate 10 nmol of total dNTPs into acid-precipitable DNA in 30 min at 74 °C

**Concentration:** 4 units/reaction (50 µl reaction volume)

**Storage:** -20 °C

Shipped in wet ice

Cat. No.	Product Description	Quantity
D1691	KlenTaq DV ReadyMix	20 reactions 100 reactions
D1816	RED KlenTaq DV ReadyMix	20 reactions 100 reactions