5x Omni Klentaq LA PCR Kit

Amount: 125 µl Omni Klentaq LA

2 x 1.25 ml tubes of 5x KM-PCR-Mix (sufficient for 500 x 25 μl reactions)

Shipping conditions: Ice Pack **Storage conditions:** -20°C F

Thermostability: Retains at least 85% activity after 1 hour at 95°C

Shelf life: At least 1 year from date of receipt under proper storage conditions.

PRODUCT DESCRIPTION:

Our PCR kit contains 5X concentrated master mix (5x KM-PCR Mix) lacking only the Omni Klentaq LA enzyme. The enzyme is provided in a separate vial, which allows an adjustment of its final concentration in PCR.

Cat #: 370

POLYMERASE TECHNOLOGY

Omni Klentaq LA is a mutant of Klentaq DNA polymerase known to be resistant to soil, blood, and other PCR inhibitors. The Long-Accurate feature allows for amplification of longer products with higher fidelity and accuracy. This kit can be used for conventional as well as real-time PCR. For real-time applications you may need to add a fluorescent dye as an alternative to probes. LA enzymes are not recommended for use with dUTP. 5x KM-PCR-Mix composition is: 250 mM Tris-Cl pH 9.2, 80 mM ammonium sulfate, 0.13% Brij 58, 17.5 mM Magnesium Chloride, and 1 mM each dNTP.

TYPICAL PCR PROTOCOL for a 25 ul reaction:

Reagent	Volume	Final Concentration
5x KM-PCR-Mix	5 μ1	1x
Left Primer	variable	200 nM
Right Primer	variable	200 nM
DNA template†	variable	0.1-100 ng
PCR Enhancer Cocktail (recommended)*	12.5 μl	1x
Omni Klentaq LA	0.05 – 0.25 μl **	
De-ionized distilled H2O	Adjust final volume to 25 ul	

 $[\]dagger$ DNA amount depends mostly on genome size and target gene copy number.

CYCLING CONDITIONS:

1. Denaturing: 94° for 2-8 minutes for 1 cycle *

2. Denaturing: 94° for 40-60 seconds

3. Annealing: 50°-68° depending on the specific Tm primers for 40-60 seconds

4. Extension: 68° for 2 min/kb target

5. Repeat steps 2-4 for 25-40 cycles

Initial 2-8 min heating step is recommended for crude samples containing 5-10% whole blood, plasma or serum.

Please visit us on the web at www.klentaq.com for troubleshooting and detailed protocols.

^{*} For optimal performance, we recommend using one of our PCR Enhancer Cocktails (PEC-1, PEC-1GC, PEC-2, or PEC-2-GC) which are specially formulated for use with whole blood, serum or plasma.

^{**} To determine specific optimal enzyme concentration, we strongly recommend an enzyme titration test for each target. Good starting amount of the enzyme per 25 ul reaction is 0.05 ul for purified DNA templates and 0.25 ul for crude samples containing 5-10% whole blood, plasma or serum. Targets larger than 1 kb may require more enzyme.

REFERENCES:

Kermekchiev, M.B., et al. (2003) Cold-sensitive mutants of Taq DNA polymerase provide a hot start for PCR. Nucl Acids Res. 31, 6139-6147.

Kermekchiev, M.B. et al. (2009) Mutants of Taq DNA polymerase resistant to PCR inhibitors allow DNA amplification from whole blood and crude soil samples. Nucl. Acids Res., 37 (5):e40 E pub.