

5x Klentaq LA PCR Kit

Cat #: 130



Amount: 125 µl enzyme and two 1.25 ml tubes of 5x Mastermix
(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 5x ready-to-use PCR kit contains Klentaq LA, an enzyme mixture which allows for longer and more accurate amplification. 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 Mastermix composition is: 250 mM Tris-Cl pH 9.2, 80 mM ammonium sulfate, 0.25% 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 Mastermix	5 µl	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
Klentaq LA	0.05 – 0.25 µl **	
De-ionized distilled H2O	Adjust final volume to 25 ul	

† DNA amount depends mostly on genome size and target gene copy number.

* For optimal performance, we recommend using one of our PCR Enhancer Cocktails (PEC-1, PEC-1GC, PEC-2, or PEC-2-GC) or 1.3 M Betaine, a general PCR enhancer.

** 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. Targets larger than 1 kb may require more enzyme.

CYCLING CONDITIONS:

1. Denaturing: 94° for 2 minutes for 1 cycle
2. Denaturing: 94° for 40-60 seconds
3. Annealing: 50°-68° depending on the specific primers (5° less than T_m) for 40-60 seconds
4. Extension: 68° for 2 min/kb target
5. Repeat steps 2-4 for 25-40 cycles

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

REFERENCES:

Barnes, W.M. (1994) PCR amplification of up to 35 kb DNA with high fidelity and high yield from bacteriophage templates, PNAS 91, 2216-2220.

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