

5x OmniTaq 2 LA PCR/RT-PCR Kit Cat #: 332



Amount: 125 µl OmniTaq 2 LA
2 x 1.25 ml tubes of 5x TM-PCR-Mix
(sufficient for 500 x 25 µl reactions)

Shipping conditions: Ice Pack

Storage conditions: For best performance, store at -20°C

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

Shelf life: At least 1 year if stored at -20°C and 10 freeze/thaws or at least 1 month if stored at 4°C.

PRODUCT DESCRIPTION:

Our PCR kit contains 5X concentrated master mix (5x TM-PCR Mix) lacking only the OmniTaq 2 LA enzyme. The enzyme is provided in a separate vial, which allows an adjustment of its final concentration in PCR.

OmniTaq 2 LA is a DNA polymerase mixture containing OmniTaq 2, a mutant of Taq DNA polymerase that provides strand-displacement and reverse transcriptase activity. It can be used as the sole enzyme in RT-PCR and RT-LAMP assays. In addition, this enzyme provides 2-3x faster PCR and some inhibition-resistance. 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 reactions you may need to add a fluorescent dye as an alternative to probes. LA enzymes are not recommended for use with dUTP. This kit is not recommended for RT-LAMP, as the buffer and dNTP concentration are incorrect. Please contact us to discuss kit options for RT-LAMP. 5x TM-PCR-Mix composition is: 250 mM Tris-Cl, 80 mM ammonium sulfate, 0.13% Brij 58, 12.5 mM Magnesium Chloride and 1 mM each dNTP. Final pH is 9.1.

TYPICAL PCR PROTOCOL for a 25 ul reaction:

Reagent	Volume	Final Concentration
5x TM-PCR-Mix	5 µl	1x
Left Primer	variable	200 nM
Right Primer	variable	200 nM
DNA template†	variable	Up to 1 ng/ul
PCR Enhancer Cocktail (optional)*	12.5 µl	1x
OmniTaq 2 LA enzyme	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.

* If inhibition-resistance is needed, 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.

CYCLING CONDITIONS FOR PCR:

1. Initial Denaturing: 94° for 2-8 minutes recommended for crude samples containing 5-10% whole blood, plasma or serum.
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 1 min/kb target
5. Repeat steps 2-4 for 25-40 cycles

CYCLING CONDITIONS FOR RT-PCR:

RT: 1. 75° for 2-8 minutes. Some highly folded RNA templates may benefit from an initial 30 seconds at 94°.
2. 68° for 30 minutes

PCR:

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

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

REFERENCE:

Barnes, W. M., et al. (2021) A Single Amino Acid Change to Taq DNA Polymerase Enables Faster PCR, Reverse Transcription and Strand-Displacement. *Frontiers in Bioengineering and Biotechnology*. 8:553474.
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