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.
Expiration: On tube label

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 µl 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 or 1.3M Betaine, a generic PCR enhancer.
** To determine specific optimal enzyme concentration, we strongly recommend an enzyme titration test for each target. A good starting amount of the enzyme per 25 µl reaction is 0.05 µl for purified DNA templates and 0.25 µl 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 Tm 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. doi: 10.3389/fbioe.2020.553474 https://doi.org/10.3389/fbioe.2020.553474