5x OmniTaq 3 PCR Kit

Amount: 125 µl OmniTaq 3 Enzyme

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 3 enzyme. The enzyme is provided in a separate vial, which allows an adjustment of its final concentration in PCR.

Cat #: 323

OmniTaq 3 is a mutant of Taq Polymerase showing enhanced performance. It can be used for conventional as well as realtime PCR. For real-time reactions you may need to add a fluorescent dye as an alternative to probes. 5x TM-PCR-Mix composition is: 250 mM Tris-Cl, 80 mM ammonium sulfate, 0.13% Brij 58, 1 mM each dNTP, and 12.5 mM magnesium chloride. 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	0.1-100 ng
PCR Enhancer Cocktail (recommended)*	12.5 μl	1x
OmniTaq 3	0.05 – 0.25 μl **	
De-ionized distilled H2O	Adjust final volume to 25 μl	

[†] 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.

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.

POLYMERASE TECHNOLOGY

^{*} 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 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 μ l reaction is 0.05 μ l for purified DNA templates and 0.25 μ l for crude samples containing 5-10% whole blood, plasma or serum. Targets larger than 1 kb may require more enzyme.