

Hot Start Enzyme Combo LA

Cat #: HS510



Amount: Hot Start Klentaq LA: 25 μ l (0.05- 0.5 μ l/ 25 μ l rxn)
Hot Start OmniTaq LA: 25 μ l (0.05- 0.5 μ l/ 25 μ l rxn)
Hot Start OmniTaq 2 LA: 25 μ l (0.05- 0.5 μ l/ 25 μ l rxn)
Hot Start OmniTaq 3 LA: 25 μ l (0.05- 0.5 μ l/ 25 μ l rxn)
Hot Start Omni Klentaq LA: 25 μ l (0.05- 0.5 μ l/ 25 μ l rxn)
Hot Start Omni Klentaq 2 LA: 25 μ l (0.05- 0.5 μ l/ 25 μ l rxn)
Hot Start Cesium Klentaq AC LA: 25 μ l (0.05- 0.5 μ l/ 25 μ l rxn)
Hot Start Cesium Klentaq C LA: 25 μ l (0.05- 0.5 μ l/ 25 μ l rxn)
Hot Start CesiumTaq LA: 25 μ l (0.05- 0.5 μ l/ 25 μ l rxn)

10x Klentaq1 Reaction Buffer (1.5 ml) for use with Hot Start Klentaq LA
10x Klentaq Mutant Reaction Buffer (1.5 ml) for use with Hot Start Omni Klentaq LA,
Hot Start Omni Klentaq 2 LA, Hot Start Cesium Klentaq AC LA, and Hot Start Cesium Klentaq C LA
10x Taq Mutant Reaction Buffer (1.5 ml) for use with Hot Start OmniTaq LA, Hot Start OmniTaq 2 LA,
Hot Start OmniTaq 3 LA, and Hot Start CesiumTaq LA
10x ZipTaq Reaction Buffer (1.5 ml) for use with Hot Start ZipTaq LA

Shipping conditions: Ambient temperature

Storage conditions: -20°C

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

Expiration: On tube label

PRODUCT DESCRIPTION:

Our Hot Start Enzyme Combo LA allows you to test 8 of our enzymes to see which one works best for your application.

10x Klentaq1 Reaction Buffer is provided for Hot Start Klentaq LA. Buffer composition is: 500 mM Tris, 160 mM ammonium sulfate, 0.5% Brij 58, and 35 mM magnesium chloride. Final pH is 9.2. pH 7.9 available upon request for better fidelity.

10x Klentaq Mutant Reaction Buffer is provided for Hot Start Omni Klentaq LA, Hot Start Omni Klentaq 2 LA, Hot Start Cesium Klentaq AC LA, and Hot Start Cesium Klentaq C LA. Buffer composition is: 500 mM Tris, 160 mM ammonium sulfate, 0.25% Brij 58, and 35 mM magnesium chloride. Final pH is 9.2. pH 7.9 available upon request for better fidelity.

10x Taq Mutant Reaction Buffer is provided for Hot Start OmniTaq LA, Hot Start OmniTaq 2 LA, Hot Start OmniTaq 3 LA, and Hot Start CesiumTaq LA. Buffer composition is: 500 mM Tris-Cl, 160 mM ammonium sulfate, 0.25% Brij 58, and 25 mM magnesium chloride. Final pH is 9.1. pH 7.9 available upon request for better fidelity.

10x ZipTaq Reaction Buffer is provided for Hot Start ZipTaq. Buffer composition is proprietary.

TYPICAL PCR PROTOCOL for a 25 μ l reaction:

| Reagent | Volume | Final Concentration |
|---------------------------------------|-----------------------------------|---------------------|
| 10x appropriate buffer (see above) | 2.5 μ l | 1x |
| dNTP mix (10 mM each) | 0.5 μ l | 200 μ M each |
| Left Primer | variable | 200 nM |
| Right Primer | variable | 200 nM |
| DNA template† | variable | 0.1-100 ng |
| PCR Enhancer Cocktail (recommended)* | 12.5 μ l | 1x |
| DNA Polymerase | 0.05 – 0.25 μ l ** | |
| De-ionized distilled H ₂ O | Adjust final volume to 25 μ l | |

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

* Our PCR Enhancer Cocktails and 1.3M Betaine (sold separately) confer additional inhibition resistance when using whole blood, serum, plasma, soil, and some inhibitory foods.

** To determine specific optimal enzyme concentration, we strongly recommend an enzyme titration test for each target. Optimal concentrations start at 0.05 μ l / 25 μ l rxn when using purified DNA template. Our Omni enzymes require more enzyme (up to 0.5 μ l / 25 μ l rxn) for use with crude samples containing 5% or more whole blood, plasma or serum, or crude soil extracts, or food matrices. For all our enzymes, targets larger than 1 kb require more enzyme.

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 T_m primers for 40-60 seconds*
4. Extension: 68° for 2 min/kb target*
5. Repeat steps 2-4 for 25-40 cycles

*Hot Start ZipTaq LA requires only 1-5 seconds for each of the denature, anneal (optional), and extension steps. For longer products, add an additional extension of 1-5 seconds per kb.

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.