# Hot Start Klentaq1

Cat #: HS100



Amount: 100 µl enyzme (sufficient for 2000 x 25 µl reactions up to 1 kb)

**Shipping conditions:** Ambient **Storage conditions:** -20°C

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:

Hot Start Klentaq1 is made with aptamer-based technology, enabling room temperature reaction set-up.

Klentaq1 is a 5'-exonuclease deficient Taq polymerase (an N-terminal deletion of Taq) with improved fidelity and thermostability.

The aptamer binds to the polymerase at sub-cycling temperatures, inactivating the enzyme and preventing spurious amplification. 10x buffer composition is: 500 mM Tris-Cl pH 9.2, 160 mM ammonium sulfate, 0.5% Brij 58, and 35 mM magnesium chloride. We also offer (upon request) 10x buffer at pH 7.9 for better fidelity.

#### TYPICAL PCR PROTOCOL for a 25 ul reaction:

Reagent	Volume	Final Concentration
10x Klentaq1 Reaction Buffer	2.5 µl	1x
DNTP mix (10 mM)	0.5 ul	200 uM each
Left Primer	variable	200 nM
Right Primer	variable	200 nM
DNA template†	variable	0.1-100 ng
Betaine 5M*	6.5 µl (optional)	1.3M
Hot Start Klentaq1**	0.05 – 0.25 μl **	2.5 units
De-ionized distilled H <sub>2</sub> O	Adjust final volume to 25 ul	-

<sup>†</sup> DNA amount depends mostly on genome size and target gene copy number.

## **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 Tm) 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.

U.S. Patent No. 5,436,149

<sup>\*</sup> Betaine is a general PCR enhancer. It usually improves the yield and specificity of amplification especially for longer targets.

<sup>\*\*</sup> To determine specific optimal enzyme concentration, we strongly recommend an enzyme titration test for each target. Targets larger than 1 kb may require more enzyme or may benefit from the LA (Long-Accurate) version of the polymerase