

## 5x Hot Start Cesium Klentaq C PCR Kit

Cat #: HS285



**Amount:** 125  $\mu$ l Enzyme  
2 x 1.25 ml tubes of 5x KM-PCR-Mix  
(sufficient for 500 x 25  $\mu$ l 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 KM-PCR Mix) lacking only the Hot Start Cesium Klentaq C enzyme. The enzyme is provided in a separate vial, which allows an adjustment of its final concentration in PCR.

Cesium Klentaq C is a cold-sensitive mutant of Klentaq1. Due to its suppressed activity at low temperatures this enzyme is designed for hot start PCR performance. It is made with aptamer-based technology further enhancing its hot start performance. This kit can be used for conventional, as well as real-time PCR. For real-time applications you may need to add a fluorescent dye as an alternative to probes. 5x KM-PCR-Mix composition is: 250 mM Tris-Cl pH 9.2, 80 mM ammonium sulfate, 0.125% Brij 58, 17.5 mM magnesium chloride, and 1 mM each dNTP,

### TYPICAL PCR PROTOCOL for a 25 $\mu$ l reaction:

Reagent	Volume	Final Concentration
5x KM-PCR-Mix	5 $\mu$ l	1x
Left Primer	variable	200 nM
Right Primer	variable	200 nM
DNA template†	variable	0.1-100 ng
PCR Enhancer Cocktail (recommended)*	12.5 $\mu$ l	1x
Hot Start Cesium Klentaq C	0.05 – 0.25 $\mu$ l **	
De-ionized distilled H2O	Adjust final volume to 25 $\mu$ l	

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

\* 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.

\*\* To determine specific optimal enzyme concentration, we strongly recommend an enzyme titration test for each target. Good starting amount of the enzyme per 25  $\mu$ l reaction is 0.05  $\mu$ l for purified DNA templates and 0.25  $\mu$ l for crude samples containing 5-10% whole blood, plasma or serum. Targets larger than 1 kb may require more enzyme or may benefit from the LA (Long-Accurate) version of the kit.

### 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](http://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.