5x CesiumTaq LA PCR Kit

Amount: 125 µl CesiumTaq 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 CesiumTaq LA enzyme. The enzyme is provided in a separate vial, which allows an adjustment of its final concentration in PCR.

Cat #: 230

CesiumTaq LA is a cold-sensitive double mutant of Taq polymerase with the Long-Accurate feature that allows amplification of longer products with higher fidelity and accuracy. Due to its suppressed activity at low temperatures this enzyme is designed for hot-start PCR 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. LA enzymes are not recommended for use with dUTP. 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:

| 111 ICALI CK I KOTOCOLIOI a 25 µi ICacioii. | | |
|---|------------------------------|----------------------------|
| Reagent | Volume | Final Concentration |
| 5x TM-PCR-Mix | 5 μ1 | 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 |
| CesiumTaq LA | 0.05 – 0.25 μ1 ** | |
| De-ionized distilled H2O | Adjust final volume to 25 µl | |

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

CYCLING CONDITIONS:

Denaturing: 94° for 2 minutes for 1 cycle
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

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.



^{*} For optimal performance, we recommend using one of our PCR Enhancer Cocktails (PEC-1, PEC-1GC, PEC-2, or PEC-2-GC) or 1.3 M Betaine, which is a general PCR enhancer.

^{**} 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.