

CleanAmp™ GC-Rich PCR 2X Master Mix Catalog # L-5102

L-5102-100 (100 reactions) L-5102-BK (Bulk amount)

CleanAmp™ GC-Rich PCR 2X Master Mix is an optimized, ready-touse mix specifically designed for robust amplification of targets over 60% in GC content. It contains CleanAmp™ dNTPs with CleanAmp™ 7-deaza-dGTP and Taq DNA Polymerase in reaction buffer. Simply add primers, template DNA and water.

QC Analysis

Functional Assay; Pass

Tested in standardized PCR assay for efficiency and specificity.

Handling & Use

Store at -20°C

Stable to 10 freeze-thaw cycles. Exposure to ambient temperatures during shipping does not adversely affect product performance.

CleanAmp™ Products: Patent Pending | RESEARCH LICENSE AGREEMENT

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Protocols

Endpoint PCR (25 µL)

- 1. Thaw CleanAmp™ GC-Rich PCR 2X Master Mix, primers and DNA template and place on ice.
 - Note: Do not vortex CleanAmp™ GC-Rich PCR 2X Master Mix. Mix thoroughly by pipetting up and down and collect by pulse centrifugation.
- 2. Prepare a reaction mixture containing all components except for the DNA template. Add CleanAmp™ GC-Rich PCR 2X Master Mix, primers and sterile de-ionized water as shown in Table 1 into thinwalled PCR tubes. Keep on ice.
- 3. Mix the reaction mixture gently to protect the enzyme, by pipetting up and down. Do not vortex. Pulse spin if necessary.
- 4. Add the appropriate volume of template DNA to reach a reaction volume of 25 μL.
- 5. Pulse spin to remove bubbles and collect reaction solution at bottom of PCR tube.
- 6. Place the tubes into a thermal cycler with a heated lid and perform the appropriate cycling conditions for standard thermal

95°C for 10 min [95°C for 40 sec; 48-60°C¹ for 1 sec²; 72°C for 1 min³] 35-40 cycles, 72°C for 7 min

7. Analyze an aliquot of the completed reaction by agarose gel electrophoresis.

Table 1

Component	Final Concentration (25 µL reaction)	Volume per reaction
CleanAmp™ GC-Rich PCR 2X Master Mix	1X	12.5 μL
Forward/Reverse Primer	50-500 nM	Variable
DNA Template ⁴	Variable	Variable
Sterile De-ionized Water	Up to 25 μL	Up to 25 μL
Total Volume (µL)	25 μL	25 μL

- The annealing temperature should be chosen for optimal PCR performance. Most primer design software recommends an annealing temperature. The annealing temperature can also be optimized experimentally by using a thermal cycler with gradient functionality or by performing sequential experiments in which the annealing temperature is varied.
- Annealing time varies between thermal cyclers. For traditional thermal cyclers, a short, second annealing time provides the best specificity. For fast cyclers use a 10 second annealing time.
- The extension time at 72 °C is recommended to be 30-60 seconds per kb of target.
- $^4\,$ 0.2 ng/µL of Human Genomic DNA is used in control reactions.