

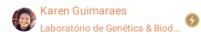


# Polymerase Chain Reaction (PCR) - DNA barcoding

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dx.doi.org/10.17504/protocols.io.vwae7ae



### PROTOCOL STATUS

#### Working

We use this protocol in our group and it is working

#### **GUIDELINES**

PCR conditions: 95°C (2min), 35 cycles of 94°C (30sec), 54°C (30sec) and 72°C (1min), followed by 72°C (10min).

BEFORE START

The reactions in 25  $\mu$ L final volume.

- 1. Add 15  $\mu$ L sterile H2O, 2.8  $\mu$ L dNTP mix (1.25 mM);
- 2. Add 2.5  $\mu$ L buffer 10X (200 mM Tris-HCl (pH = 8,4) + 500 mM KCl);
- 3. Add 2.5 µL de MgCl2 (50 mM);
- 4. Add 0.5 μL of each primer (5μM);
- 5. Add 0.2  $\mu$ L Taq DNA polymerase (5U/ $\mu$ L) and 1  $\mu$ L of genomic DNA (100ng/ $\mu$ L)

## MATERIALS TEXT

Sterile H2O; dNTP mix (1.25 mM); buffer 10X (200 mM Tris-HCl (pH = 8,4) + 500 mM KCl); MgCl2 (50 mM); Primer Fish F1 and Fish R1 (5 $\mu$ M); Taq DNA polymerase.

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- 5. Add 0.2  $\mu L$  Taq DNA polymerase (5U/ $\mu L)$  and 1  $\mu L$  of genomic DNA (100ng/ $\mu L)$

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