

★ Storage

Stable at -20°C in a constant temperature freezer.

✎ Contents

- Product manual
- **amfiEco** Taq DNA Polymerase, 1u/ul
- **amfiEco** 2X Reaction Buffer

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★ Introduction

amfiEco Taq DNA Polymerase is a thermostable DNA polymerase that possesses a 5'→3' polymerase activity and a double-strand specific 5'→3' exonuclease activity. 2X PCR Master buffer including dNTPs and Mg⁺⁺ minimises reagents handling steps and reduces the risk of contamination. For added convenience, **amfiEco** Taq DNA Polymerase contains red and yellow loading dyes to allow loading of PCR product directly on a gel after thermal cycling, minimizing pipetting steps and providing easy visualization of sample. The red dye runs in a range between 500bp(2% gel) and 1500bp(0.8% gel) and the yellow dye runs at less than 10bp.

★ Components for each reaction

1u/rxn **amfiEco** Taq DNA Polymerase, Reaction Buffer, 1.75mM MgCl₂, 0.2mM dGTP, 0.2mM dATP, 0.2mM dTTP, 0.2mM dCTP, stabilizers, and red and yellow loading dyes.

★ Unit Definition

One unit is defined as the amount of the enzyme required to catalyze the incorporation of 10nmol of dNTPs into an acid insoluble form in 30 min at 72°C.

★ Quality Control

PCR, Activity, endonuclease/nickage, Specific performance test.

★ Recommendation

1. Since PCR is a powerful technique capable of amplifying trace amounts of DNA, all appropriate precautions should be taken to avoid cross-contamination.
2. Use of "clean" dedicated automatic pipettors and aerosol resistant barrier tips are recommended.
3. MgCl₂ is included in the 2X Reaction Buffer at a final concentration of 1.75 mM, which is sufficient for most targets. For some targets, more Mg²⁺ may be required.
4. Always perform control reactions without template DNA to check for the absence of contamination.

★ Protocol

1. Gently vortex and briefly centrifuge all solutions after thawing, keep it all components on ice.
2. Add the following components to a thin-wall tube sitting on ice.

Component	Volume per 25ul of rxn	Final Conc.
amfiEco 2X Reaction Buffer	12.5ul	1X
Forward primer, 10uM	0.25-2.5ul	0.1-1.0uM
Reverse primer, 10uM	0.25-2.5ul	0.1-1.0uM
DNA template	1-5ul	< 1ug
amfiEco Taq DNA Polymerase	1ul	1U
Nuclease free water to	25ul	N.A

3. Gently vortex and spin down to collect drops.
4. When using the thermal cycler without a heated lid, overlay the reaction mixture with one-half volume of mineral oil.
5. Perform 25-40 cycles of PCR amplification.

Note : Cycling Parameters

Segment	Number of Cycles	Temperature	Duration
1	1	94°C	2min
2	25-40	94°C	2min
		Primer Tm-5°C	2min
		72°C	1min per kb
3	1	72°C	10min

This Cycling Parameter serves as a guideline for PCR amplification. Optimal reaction condition such as PCR cycles, annealing temperature, extension temperature, and predenaturation time and temperature may vary and must be individually determined.

★ Related Product

Product Name	Cat No
amfiSure Taq DNA Polymerase	P0310
amfiSure PCR Premix, individual 0.2ml tube	P0313
amfiSure PCR Premix, custom order	P0318