

2X Thermo-Start™ High Performance ReddyMix™ PCR Master Mix

Description: Thermo-Start™ *Taq* DNA Polymerase is a chemically modified version of ThermoPrime *Taq* DNA Polymerase. It is completely inactive at room temperature, preventing the formation and subsequent amplification of non-specific products. The enzyme requires an **activation step at 95°C for 15 minutes**.

ReddyMix™ PCR Master Mix is a ready-to-use master mix. It is a convenient way of amplifying DNA fragments without the need to thaw individual components, reducing the risk of contamination and pipetting errors. The Thermo-Start™ *Taq* DNA Polymerase, dNTPs, reaction buffer and magnesium chloride are all present in the mix. ReddyMix™ Master Mix also contains a dye and precipitant to facilitate gel loading.

Enzyme Source: *Thermus aquaticus*

Associated Activities: Thermo-Start™ has 5' to 3' polymerization and exonuclease activity but lacks 3' to 5' exonuclease activity (proofreading).

Kit Contents

| Vial (cap color) | Pack Size | |
|--|-----------|----------|
| | A | B |
| 2X Thermo-Start HP ReddyMix Master Mix (clear) | 2 x 1ml | 20 x 1ml |

Master Mix: Each vial contains 1ml of a 2X working concentration PCR Master Mix sufficient for 80 x 25µl reactions. The final reaction 1X contains:

| | |
|---|---|
| 0.625 units | Thermo-Start™ <i>Taq</i> DNA Polymerase |
| 1X | Thermo-Start High Performance Reaction Buffer |
| 1.5mM | MgCl ₂ |
| 0.2mM | each of dATP, dCTP, dGTP and dTTP |
| Precipitant and red dye for electrophoresis | |

Storage Conditions: Store ReddyMix at -20°C until ready for use for up to 1 year. Avoid freeze thawing. Vials can be stored at 4°C for up to 1 month. Shipped on ice within the UK and on dry ice internationally and within the US.

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Example of Protocol:

Mix gently and spin down the master mix prior to use

| | Volume | Final Concentration 1X |
|--|----------|------------------------|
| 2X Thermo-Start HP ReddyMix Master Mix | 12.5µl | 1X |
| Primer forward (12.5µM each) | 1µl* | 0.5µM* |
| Primer reverse (12.5µM each) | 1µl* | 0.5µM* |
| Water (PCR Grade) | Variable | |
| DNA Template | 0.5µl* | 0.5 - 125ng* |
| Total volume | 25µl | |

*Scale up or down the volume and concentration as appropriate

Tip:

The gel precipitant in ReddyMix™ Master Mix causes a slight increase in the thermal mass of the reaction mix. In a small number of cases this may necessitate some minor re-optimization of the thermal cycler program. If this is the case we suggest increasing the temperature of the denaturation step by 1–2°C and decreasing the temperature of the annealing step by 1–2°C. Alternatively, increase the duration of each step by 5-10 seconds.

Example of Program:

| | Temp. | Time | Number of cycle |
|-----------------------------|-------------|---------------|-----------------|
| Initial Denaturation | 95°C | 15 min | 1 cycle |
| Denaturation | 95°C | 25 sec | 30 to 40 cycles |
| Annealing | 48-63°C | 35 sec | |
| Extension** | 72°C | 65 sec | |
| Final Extension | 72°C | 5 min | 1 cycle |

**Increase length of time in proportion to size of amplicon, *Taq* DNA Polymerase extends at approximately 1000 bp/min.

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Ordering Information:

| | | |
|-----------|--|------------------|
| AB-1962/A | 2X Thermo-Start™ HP ReddyMix™ PCR Master Mix | 160 x 25µl rxns |
| AB-1962/B | 2X Thermo-Start™ HP ReddyMix™ PCR Master Mix | 1600 x 25µl rxns |

Troubleshooting

For troubleshooting, see www.abgene.com/troubleshoot.asp or contact Thermo Fisher Scientific (ABgene) Tech Support at abgene.techsupport@thermofisher.com

UK Tech Support, call +44 (0) 1372 840 410

For all other regions, please contact your local Thermo Fisher Scientific (ABgene) office / distributor.

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