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PCRBIO Taq Mix Red

Product description

PCRBIO Taq Mix Red uses the latest developments in polymerase technology and buffer chemistry to enhance PCR speed, yield and specificity. The enzyme and buffer system allow for superior PCR performance on complex templates such as mammalian genomic DNA. Reactions can be directly loaded onto agarose gels without additional loading buffer.

PCRBIO Taq Mix Red is a robust mix for all your everyday PCR applications including genotyping, screening and library construction. This 2x mix contains PCRBIO Taq DNA Polymerase which performs consistently well on a broad range of templates (including both GC and AT-rich).

PCRBIO Taq DNA Polymerase has an error rate of approximately 1 error per 2.0×10^5 nucleotides incorporated. PCR products generated with PCRBIO Taq DNA Polymerase are A-tailed and may be cloned into TA cloning vectors.

High-throughput screening has resulted in a buffer system that allows efficient amplification from GC-rich and AT-rich templates, under fast and standard cycling conditions.

Component	200 reactions	1000 reactions
2x PCRBIO Taq Mix Red	5 x 1mL	25 x 1mL

Shipping and storage

On arrival the kit should be stored at between -30°C and -15°C. Avoid prolonged exposure to light. If stored correctly the kit will retain full activity for 12 months. The kit can be stored at 4°C for 1 month. The kit can go through 30 freeze/thaw cycles with no loss of activity.

Limitations of product use

The product may be used for in vitro research purposes only.

Technical support

Help and support is available on our website at https://pcrbio.com/resources/ including answers to frequently asked technical questions. For technical support and troubleshooting you can submit a technical enquiry online, or alternatively email technical@pcrbio.com with the following information:

- Amplicon size
- Reaction setup
- Cycling conditions
- Screen grabs of gel images

Important considerations

2x PCRBIO Taq Mix Red: The 2x mix contains PCRBIO Taq DNA Polymerase, 6mM $MgCl_2$, 2mM dNTPs, enhancers, stabilizers and a red dye for tracking during agarose electrophoresis. It is not recommended to add further PCR enhancers or $MgCl_2$ to the reaction. The buffer composition has been optimised to maximise PCR success rates.

Template: For eukaryotic DNA use between 5ng and 500ng per reaction, for cDNA use below 100ng per reaction.

Primers: Primers should have a predicted melting temperature of around 60˚C, using default Primer 3 settings (http://bioinfo.ut.ee/primer3/). The final primer concentration in the reaction should be between 0.2µM and 0.6µM.

Annealing: We recommend performing a temperature gradient to experimentally determine the optimal annealing temperature. Alternatively, we recommend a 55˚C annealing temperature then increase in 2˚C increments if non-specific products are present.

Extension: Optimal extension is achieved at 72˚C. The optimal extension time is dependent on amplicon length and complexity of template. 20 seconds per kilobase (kb) is recommended for amplification from eukaryotic DNA for amplicons between 1kb and 6kb. For shorter amplicons, faster cycling is possible.

Agarose gel electrophoresis dye migration: The 2x mix contains a red dye for tracking during agarose gel electrophoresis. In a 2% agarose TAE gel the dye migrates at a rate equivalent to 350bp of DNA. In a 1% agarose TAE gel the dye migration rate is equivalent to 600bp of DNA.

Reaction setup

1. Prepare a master mix based on the following table:

Reagent	50µL reaction	Final concentration	Notes
2x PCRBIO Taq Mix Red	25.0µL	1x	
Forward primer (10µM)	2.0µL	400nM	See above for optimal primer design
Reverse primer (10µM)	2.0µL	400nM	
Template DNA	<100ng cDNA, <500ng genomic	variable	See above for template considerations
PCR grade dH_2O	Up to 50µL final volume		

2. Cycle using conditions based on the following table:

Cycles	Temperature	Time	Notes
1	95˚C	1min	Initial denaturation
40	95˚C	15 seconds	Denaturation
	55˚C to 65˚C	15 seconds	Anneal
	72˚C	1 to 120 seconds	Extension (20 seconds per kb)