vivantis

DNA AMPLIFICATION PRODUCT

2X ViRed *Pfu* Master Mix

Product Datasheet

Product No : CLMM03
Quantity : 100 reactions

2X ViRed PMM

Lot :
Expiry Date :
Supplied with : 4 x 625µl **2X ViRed *Pfu* Master Mix***
3ml of Nuclease-free Water

Store at -20°C

*2X ViRed *Pfu* Master Mix consists of *Pfu* DNA Polymerase, 2X ViBuffer *Pfu*, dNTPs, inert red dye and stabilizers.

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Description :

2X ViRed *Pfu* Master Mix is an optimized ready-to-use 2X concentrated DNA amplification mixture premixed with red color tracking dye. The ViRed *Pfu* Master Mix contains *Pfu* DNA Polymerase, reaction buffer, dNTPs, inert red dye and stabilizers needed for routine DNA amplification to obtain a wide range of PCR and DNA products up to 8kb. An inert red dye and stabilizers allows direct loading of final products onto gels for electrophoresis. The red color dye migrates at approximately 400bp on 1% agarose in 1X TBE Buffer. *Pfu* DNA Polymerase is an extremely thermostable proofreading DNA polymerase. It exhibits the 3' to 5' proofreading activity, resulting in over 10-fold higher fidelity than possible with *Taq* DNA Polymerases.

Features:

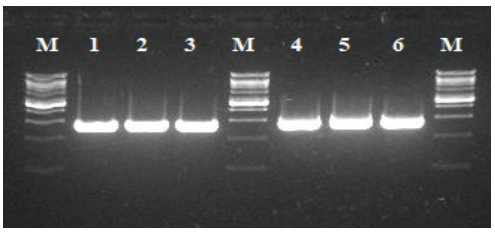
- Suitable for all routine DNA amplification applications
- Recommended for use in high-fidelity amplification, amplification of GC-rich sequences or problematic secondary structures, primer extension reactions at evelated temperatures and cloning of blunt-ended amplification products.
- Reduces set-up time and buffer-dye mixing
- No additional loading dye needed – direct loading of final products onto gels

Storage and Stability:

- Stable at -20°C for 18 months or at 4°C for 6 months if properly stored
- Stable for 20 freeze-thaw cycles. To avoid frequent freeze-thaw, keeping small aliquots at -20°C is recommended
- For daily use, keeping aliquots at 4°C is recommended

Quality Control:

All preparations are assayed for contaminating endonuclease, exonuclease, and non-specific DNase activities. Functionally tested in DNA amplification.



Amplification of 1.5kb DNA fragment from pTZ DNA region using 2X ViRed *Pfu* Master Mix in a 50µl reaction mixture (1.0% TBE agarose gel).

Lane M : VC 1kb DNA Ladder
Lane 1 : DNA amplification product generated with 1u of *Pfu* DNA Polymerase
Lane 2 : DNA amplification product generated with 2X ViRed *Pfu* Master Mix (store at -20°C)
Lane 3 : DNA amplification product generated with 2X ViRed *Pfu* Master Mix (after 20 freeze-thaw cycles)
Lane M : VC 1kb DNA Ladder
Lane 4 : DNA amplification product generated with 1u of *Pfu* DNA Polymerase
Lane 5 : DNA amplification product generated with 2X *Pfu* Master Mix (store at -20°C)
Lane 6 : DNA amplification product generated with 2X *Pfu* Master Mix (after 20 freeze-thaw cycles)

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RECOMMENDED PROTOCOL FOR 2X ViRed *Pfu* Master Mix:

Gently mix all solutions after thawing. Spin down briefly and keep on ice.
Add the following components in a 0.2ml thin walled PCR tube on ice.

For 50µl reaction volume:

Reagent:	Volume	Final Concentration
2X ViRed *Pfu* Master Mix	25µl	*1X
Primers (Fwd / Rev)	Variable	0.1 - 1 µM each
DNA Template	Variable	0.02 - 5µg
Water, nuclease-free	Adjust final volume to 50µl	

*1u unit Pfu DNA polymerase, 1X ViBuffer Pfu, 0.2mM dNTPs and stabilizers.
Note : Smaller reaction volume may be achieved provided that the same final concentration of each reaction component is maintained.

CYCLING CONDITIONS (100bp-5kb)	
Denaturation	94°C for 5 minutes
Denaturation	94°C for 30 seconds
Annealing	50 - 68°C for 1 minutes
Extension / 1kb	72°C for 2 minutes
Final Extension	72°C for 7 minutes

25 - 35 cycles

This protocol may change depending on the template DNA and primers used.

Product Use Limitation
This product is for research purpose and *in vitro* use only

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