

TECHNICAL DATA SHEET

VerityMAX™ DNA Polymerase Master Mix

Catalog Numbers: 31-5031-0080R
31-5031-0400R

PRODUCT INFORMATION

Contents:

31-5031-0080R (80 rxns)
VerityMAX™ DNA Polymerase Master Mix (2X): 2 x 1.0 mL

31-5031-0400R (400 rxns)
VerityMAX™ DNA Polymerase Master Mix (2X): 10 x 1.0 mL

Use By: 6 months from date of receipt

DESCRIPTION

VerityMAX™ DNA Polymerase Master Mix contains a proprietary blend of our VerityPfu™ and Classic++™ Hot Start *Taq* DNA polymerases, engineered and optimized specifically for generating amplicons of up to 35 kb in length with 2.5x higher fidelity than standard *Taq* polymerase. VerityPfu polymerase was developed using site directed mutagenesis that allowed for the positive selection of a very high fidelity *Pfu* variant optimized for use at room temperature. Accordingly, the VerityPfu enzyme possesses exceptional 3'→5' exonuclease, or proofreading, activity. Classic++ Hot Start *Taq* DNA Polymerase is Tonbo's next generation recombinant *Taq* polymerase that possesses 5'→3' exonuclease activity. Proprietary Classic++ Hot Start mAbs (monoclonal antibodies) have been developed that specifically block polymerase activity below 70°C, allowing for convenient room temperature reaction set up. Hot start technology improves yields by minimizing or eliminating primer dimer formation and non-specific amplification.

Tonbo's VerityMAX DNA Polymerase Master Mix is ideal for a wide variety of fast or standard PCR protocols when additional speed, sensitivity, specificity and yields are desired. Use the VerityMAX Master Mix to accurately and reliably amplify DNA of up to 35 kb from complex GC/AT rich genomic DNA, plasmid templates, crude PCR and for bacterial colony PCR. The resultant amplicons are both blunt-ended and A-tailed and can be used in a wide variety of downstream applications, including TA and blunt-end cloning.

STORAGE

Store kit at -20°C upon arrival and limit exposure to light. The kit may undergo up to 30 freeze/thaw cycles without loss of activity. When stored correctly this kit will retain activity for up to 6 months. This product can be stored at 4°C for up to 1 month.

BIOLOGICAL SOURCE

VerityPfu DNA Polymerase is a single recombinant polypeptide of bacterial origin having a molecular weight of ~90 kDa, originally derived from the hyperthermophilic archaeum *Pyrococcus furiosus*. Classic++ Hot Start *Taq* DNA Polymerase is a single recombinant polypeptide of bacterial origin having a molecular weight of ~94 kDa, originally derived from the YT-1 strain of *Thermus aquaticus*. Classic++ Hot Start mAbs are of murine origin and are reactive with select epitopes found within recombinant forms of the YT-1 strain of *Thermus aquaticus*.

APPLICATION NOTES

Master Mix: VerityMAX DNA Polymerase Master Mix (2X) contains VerityPfu and Classic++ Hot Start *Taq* DNA polymerases, 6 mM MgCl₂, 2 mM dNTPs plus a proprietary mix of stabilizers and enhancers. This Master Mix has been rigorously developed for optimal PCR success. We do not recommend adding additional MgCl₂ or enhancers to the reaction mix.

Primers: We recommend that primers have a predicted melting temperature of approximately 60°C using default Primer 3 settings (<http://bioinfo.ut.ee/primer3/>). For each reaction, a final primer concentration of 0.2 - 0.6 µM is suggested.

Template: For cDNA templates, use less than 100 ng per reaction. For eukaryotic DNA templates, use 5 - 500 ng per reaction.

Annealing Temperature: It is preferable to generate a temperature gradient in order to empirically determine the optimal annealing temperature for the reaction. Otherwise, one can start with an annealing temperature of 55°C and, if non-specific products are observed, increase in 2°C increments (up to maximum 65°C) until an optimal temperature is reached.

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Extension Temperature: We observe optimal extension at 72°C. Extension time depends on both the template complexity and amplicon length. For amplicons under 5 kilobases (kb) from eukaryotic DNA, we recommend 15 seconds per kb. Increase extension time to 40-60 seconds per kb for amplicons between 5 and 35 kb.

REACTION SETUP / QUICK PROTOCOL

1. Ensure all components are thawed and mixed well. Equilibrate VerityMAX Master Mix to room temperature. Refer to Table 1 for reaction preparation. If preparing multiple reactions, assemble all common components into a master reaction mix. If working with final reaction volumes less than 50 µl, scale component volumes accordingly. Refer to Application Notes for modifications when using special sample types.
2. As applicable, transfer the recommended volume of master reaction mix, primers and sample template DNA to individual PCR tubes or plates, seal and spin briefly to mix.
3. Refer to the cycling conditions (Table 2) to perform the PCR.

Table 1. Reaction Preparation

Reagent	50 µL reaction	Final Concentration	Notes
2x VerityMAX™ Master Mix	25.0 µL	1x	
Forward Primer (10 µM)	2.0 µL	400 nM	See above for optimal primer design
Reverse Primer (10 µM)	2.0 µL	400 nM	
Template DNA	<100 ng cDNA <500 ng genomic	variable	See above for template considerations
Nuclease free dH ₂ O	Up to 50 µL final volume		

Table 2. Cycling Conditions

Cycles	Temperature	Time	Notes
1	95°C	1 minute	Initial denaturation
40	95°C	15 seconds	Denaturation
	55°C - 65°C	15 seconds	Anneal
	72°C	10 minutes	Extension (50 seconds per kb); See above for extension considerations

TECHNICAL SUPPORT

Please provide the following information to support@tonbobio.com for troubleshooting and technical support:

- Catalog and batch numbers
- Reaction set-up (master mix)
- Cycling conditions
- Amplicon size
- Screen shots of gel images
- Detailed description of the issue

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