LAMDA BIOTECH

PRODUCT INFORMATION

Catalog No.: D136G

Product Name: **HotStart PCR Master Mix in Green**

Size: 5000 µl

Description: Ready-to-use 2X hot start PCR master mix in green color, containing a *HotStart Taq DNA polymerase* and a PCR enhancing DNA polymerase. Users only need to add template, primers and H_2O for the reaction. The **HotStart PCR Master Mix** contains inert, non-toxic dyes to make the color in green, which makes it easy to visualize the PCR mixing step, and also allows direct loading of PCR products for electrophoresis.

The HotStart *Taq* DNA Polymerase is a chemically modified Taq DNA Polymerase, whose enzyme activities can only be activated after 3-5 minutes of incubation at 95°C. The HotStart Taq Polymerase uses amplification conditions for regular *Taq* DNA Polymerase, except no polymerase activity will be present before the onset of thermal cycling. This prevents nonspecific DNA amplification and primer dimer formation. The amplified products, up to 7Kb in length, contain PCR products with blunt end and 3'-overhanging A end. This allows flexible protocols for PCR cloning, if the amplified fragments need to be cloned.

Quality Testing: All the DNA Polymerases are highly purified and free of contaminating endonucleases, exonucleases and nicking activity. For endonuclease assay, 1µg of Lamda/Hind III DNA is incubated with 20 units of enzyme in assay buffer at 75°C for 16 hours with no visible contaminating activity observed. Also, every lot is tested for its performance consistency.

Storage: 4°C for up to one month, or -20°C for long term storage.

Unit Definition: One unit incorporates 10nmoles of dNTPs into acid-insoluble material in 30 minutes at 70°C.

1x Composition: 20mM KCl, 20mM Tris HCl (pH8.4), **1.5mM $MgCl_2$***, 200µM dNTPs, 2.5units/25ul of *Taq* DNA polymerase, trace amount of red dye, PCR enhancer and enzyme stabilizers.

***Magnesium Chloride:** In general, 1.5mM $MgCl_2$ is sufficient for most of the PCR experiments. However, this may vary with different conditions and primer sets. Some primers/templates may require adjustments for $MgCl_2$ concentration, which can be achieved as shown below:

Final $MgCl_2$ conc.	Additional 25mM $MgCl_2$ per 50µl reaction
1.5mM	------
2.0mM	1.0µl
2.5mM	2.0µl

Directions for use: For a 50µl reaction: use 25µl of **HotStart PCR Master Mix**; add template, primers and H_2O to a final volume of 50µl. Cycling conditions vary for different templates and primers. To start with, try 30 cycles as follows: denature at 94°C for 30sec, anneal around 55°C for 30sec, and extend at 72°C for 1 minute/kb. After the PCR cycles, extend at 72°C for another 5 minutes to complete the PCR. Then store the reaction at 4°C.

This product is for research use only