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Hot Start Mastermix & Hot Start Mastermix Red

Catalogue Numbers:

DNP-H105	100 Reactions
DNP-H205	500 Reactions
DNP-H105R	100 Reactions
DNP-H205R	500 Reactions

Features

- Same high specificity and performance Hot Start Taq
- Reduced risk of contamination
- Dramatically decreases the time required for reaction set-up

Applications

- Hot-start PCR assays
- Products suitable for TA cloning

Description

Hot Start Mastermix is a complete ready-to-use heat-activated 2x reaction-mix, which simply requires the user to add only water, template and primers, and then pre-heat to 95°C for 10 minutes to successfully carry out PCR assays. The 10-minute activation step eliminates the presence of non-specifics such as primer-dimers and mis-primed products, since the enzyme is inactive at initial low temperatures.

Hot Start Mastermix Red combines all of the features and advantages of Hot Start Mastermix, and contains an additional inert red dye. This non-toxic, non-hazardous red dye allows users to load samples directly onto a gel, without the need to add loading buffer since the mix is of sufficiently high density to sink to the bottom of the gel. Hot Start Mastermix and Hot Start Mastermix Red dramatically reduce the time needed to set up reactions, thereby reducing the risk of contamination. Greater reproducibility is ensured, by reducing the number of pipetting steps that can lead to pipetting errors.

Product Specifications

Batch details:

Batch No: See vial

Components

Hot Start Mastermix	100 Reactions	500 Reactions
Hot Start Mastermix	2 x 1.25ml	10 x 1.25ml
50mM MgCl ₂ Solution	1.2ml	1.2ml
Hot Start Mastermix Red		
Hot Start Mastermix Red	2 x 1.25ml	10 x 1.25ml
50mM MgCl ₂ Solution	1.2ml	1.2ml

Hot Start Mastermix & Hot Start Mastermix Red Protocol

Reaction Conditions (for a 50µl reaction)

Hot Start Mastermix/Mastermix Red	25ul
Template and Primers	As required
Water (ddH ₂ O)	Up to 50ul

Activation: pre-heating step at 95°C for 10 minutes

Denaturation: 94-96°C

Annealing: depends on primer T_m

Extension: 72°C (allowing 15-30 seconds/kb)

This data is intended for use as a guide only; conditions will vary from reaction to reaction and may need optimization.

The enzyme must be activated by heat treatment before PCR cycling. All reaction components should be added to the reaction, and then **pre-incubated at 95°C for 10 minutes**.

If the PCR extension time exceeds 3 minutes, program the thermal cycler to run for a maximum of 30 cycles. Increasing the number of cycles may lead to smearing of bands when the samples are run on an agarose gel.

For optimal resolution of PCR products, we recommend the use of Tris-Acetate EDTA (TAE) buffer for gel preparation and electrophoresis.

When used in a 50µl reaction volume, Hot Start Mastermix contains 3mM MgCl₂. An additional tube of 50mM MgCl₂ is provided should any fine adjustments be necessary. The table below shows the volume of MgCl₂ that must be added to a 50µl final reaction to achieve the desired final concentration.

Final MgCl ₂	MgCl ₂ to be added
3.0mM	0ul
3.5mM	0.5ul
4.0mM	1ul

Reagent Specifications:

MgCl₂ Stock Solution: 50mM MgCl₂

Storage Conditions:

Hot Start Mastermix and Hot Start Mastermix Red can be stored for up to 6 months at -20C, or up to 2 weeks at 4C. Repeated freeze/thaw cycles should be avoided.

Shipping Conditions:

On Dry Ice or Blue Ice