



2X HotSybr PCR Reaction Mix

CAT # (SKU): HSM-400RF

FOR RESEARCH USE ONLY

Storage

This product should be stored at -20°C . To avoid repeated freeze-thaw, opened vials should be kept at 4°C .

About the product

A key component of 2X HotSybr PCR Reaction Mix is a stringent hot-start DNA polymerase. This proprietary technology ensures robust and highly sensitive PCR amplification and quantification.

Primer design

Appropriate software, such as Primer Express, should be used.

1. T_m : $58\sim 60^{\circ}\text{C}$ with 17 ~ 30 nucleotides in length
2. To design a small amplicon, <150bp
3. To avoid secondary structures
4. To avoid more than 3 consecutive Gs
5. To avoid complementary 3' ends

Conditions

The recommended thermocycling conditions is: 95°C , 10 min. \rightarrow (95°C , 5 sec. \rightarrow 60°C , 30 sec.) for 40 cycles \rightarrow melting curve.

Tips on how to achieve best performance

1. Primer concentration should not be high. 100nM to 300nM each primer usually gives best results.
2. Melting curve is a useful way to distinguish non-specific amplification products and primer dimers from desired product. It is recommended to run melting curve following amplification cycles.
3. To do replicates.
4. To reduce pipetting error: preparing amplicon specific master mix, using repeat pipet, and keeping pipetting volume in manufacture suggested range are very effective to get tight Ct

This reaction mix DOES NOT contain internal passive reference dye (ROX).