SinaSYBR Green HS-qPCRMix, 2x

For Research Use Only

Cat. No.: MM2171 **Quantity:**100 reactions/ 25µl

Store at:-20°C (not more than 50 thawing-freezing cycles) **Shipment:** Wet or dry Ice

Description: SinaSYBR Green HS-qPCR Mix, 2x is developed for quantitative real-time PCR with fluorescent dye SYBR Green I. SinaSYBR Green HS-qPCR Mix, 2x includes all of components necessary to PCR (**highly processive recombinant HS-*Taq* DNA polymerase**, **deoxynucleoside triphosphate mix**, **PCR buffer**, **Mg^{2+}**, **SYBR Green I**, **Inert dye** except for DNA template and primers).

The mix is optimized for conducting consistent and efficient real-time Hot start PCR of genomic, plasmid and viral DNA samples. The solution includes substances increasing half-life and processivity of HS-*Taq* DNA polymerase by enhancing its stability during PCR. SinaSYBR Green HS-qPCR Mix, 2x contains components that influence primer annealing temperature and characteristics of template melting, thus enabling to increase the specificity of PCR and use templates with complicated partial structure. The DNA polymerase included in the mix is inactive at room temperature, its activation requires preheating at 95 °C for 5 min.

Components (supplied):

SinaSYBR Green HS-qPCR Mix, 2x	1.25 ml

SinaSYBR Green HS-qPCR Mix, 2x contains:

100mM Tris-HCl(pH8.5 at 25 °C) 100mM KCl, 0.4mM of each deoxynucleoside triphosphate,3mM $MgCl_2$,0.06U/µl *Taq*DNA polymerase,0.025% Tween20,stabilizers of HS-*Taq* DNA polymerase , SYBR Green I, and inert dye.

Applications:

- Real-Time PCR with intercalating dye SYBR Green I
- Conventional PCR
- High-throughput PCR
- Genotyping

HS- *Taq*DNA Polymerase features

Recombinant HS-*Taq*DNA polymerase has 5'→3'DNA-dependent polymerase activity and5'→3' exonuclease activity.

SYBR Green I

SBRY Green I is a fluorescent intercalating dye for quantitative and qualitative detection of PCR products during real-time PCR. SYBR Green I provide easy and economical way for detection and quantitative assessment of PCR products during real-time PCR without a need for specific fluorescent probes. During amplification, SYBR Green I dye penetrates into the minor groove of DNA products and emits stronger fluorescent signal than unbound dye. Absorption and emission maxima of SYBR Green I are 494nm and 521nm, respectively, which enables to use it for every real-time PCR platform existing to date.

Inert dye

The inert dye included in SinaSYBR Green HS-qPCR Mix, 2x does not reduces PCR efficiency; it facilitates monitoring of multi-well plate pipetting. Absorption maximum of the blue dye is 615nm.

Benefits of use

- the enzyme with hot start capability increases reaction specificity and sensitivity
- HS-*Taq* DNA polymerase activation requires not more than 5 min heating
- High selectivity and reaction yield
- The mix is colored for easy pipetting
- Reduced preparation time

•Low contamination risk when mixing PCR components

•Standardized conditions of the same-type reactions (reduced pipetting error during mixing PCR components in a series of experiments)

Limits of use

Not recommended to use for real-time PCR whit fluorescently labeled probes.

Recommended qPCR reaction mix:

1. Defrost the reaction mixture and stir thoroughly.
2. Put thin-wall PCR tubes on ice and add the following components considering the final volume of a reaction mixture equal to 25 µl:

Component	Volume	Finalconcentration
SinaSYBR Green HS-qPCR Mix, 2x	12.5	1×
Forward primer	variable	0.1–600nM
Reverse primer	variable	0.1–600nM
DNA	variable	1 pg–1µg
Sterile water	up to25µl	

Recommended qPCR cycles:

Step	Temperature, °C	Incubation time	Number of cycles
Preliminary denaturation	95	5-7min	1
Denaturation	95	15sec	25- 50
Annealing	50- 68	10-30sec	
Elongation	58- 72	30-60sec	
Melting curve (recommended)	65 - 95		1

Note: Monitoring of real-time PCR can be conducted at 72°C in case of absence of non-specific products (primer dimers). In case if non-specific products are formed whit Tm_1 lower than Tm_2 of the target product. Monitoring should be performed at the temperatures between Tm_1 and Tm_2.