ig™ SYBR Green qPCR 2X Master Mix

Description

Ig SYBR® Green qPCR 2x master mix is a ready-to-use cocktail containing all components except primers and template, for the amplification and detection of DNA in qPCR. The Ig SYBR® Green qPCR 2x master mix with integrated chemically-modified hot start Taq DNA polymerase, SYBR® Green I fluorescent dye, ROX dye, MgCl2, dNTPs and stabilizers. This master mix is ideal for high-throughput real-time PCR screening and validation. The amplification step features a high quality hot start Taq DNA Polymerase which offers higher fidelity and better amplification.

Applications

- Gene expression data validation.
- Absolute quantification
- Mutation detection
- Pathogen detection
- Viral detection
- Genetically modified organisms (GMO) characterization
- Genetic profiling

Benefits

- Enhanced efficiency, specificity, and sensitivity
- Compatible with all real-time PCR instruments
- Superior gene expression results under various cycling conditions
- Robust and active for cDNA synthesis at temperatures up to 55 °C.

Product Includes

1) ig™ SYBR Green qPCR 2x Master Mix

Storage Temperature

−20 °C

Protocol

- Place kit components, primers and RNA samples on ice.
- 2. Mix and then centrifuge briefly to collect contents at the bottom of the tube.
- 3. Prepare a master mix for each reaction and control plus 10% extra to allow for pipetting error according to the following table:

| PCR reaction set up: | | |
|----------------------------------|---------|--|
| Diluted cDNA | 1-5 µl | |
| Forward primer (5 µM) | 1.0 µl | |
| Reverse primer (5 µM) | 1.0 µl | |
| ig™SYBR Green qPCR 2x master mix | 10.0 µl | |
| H ₂ O up to | 20.0 µl | |

- 4. Mix the reaction mixture thoroughly.
- Program the thermal cycler according to the manufacturer's instructions.
- 6. A typical PCR cycling program is outlined in the following table.

| PCR Cycling Conditions: | | | | |
|-------------------------|------------------------------------|--------|--------|--|
| Steps | Temperature | Time | Cycles | |
| Initial denaturation | 95 ℃ | 15 min | 1 | |
| Denaturation | 95 °C | 5 sec | 30-40 | |
| Annealing/Extension** | ~60 °C | 30 sec | | |
| Melting curve analysis | According to instrument guidelines | | | |

- 7. Place the PCR tubes in the thermal cycler and start the cycling program.
- Analyze the data according to manufacturer protocol.

^{**} For 3 step cycling protocols, anneal at optimal annealing temperature for 30 sec followed by the minimum time required for data acquisition at 72 °C according to instrument guidelines.