



## OneStep RT-PCR Kit

| Cat. No. | size          |
|----------|---------------|
| E0803-01 | 25 reactions  |
| E0803-02 | 100 reactions |

**Storage Conditions:** Store at -20°C.

### Quality Control:

All preparations are assayed for contaminating endonucleases, exonucleases, nonspecific RNases, single- and double-stranded DNase activities.

OneStep RT-PCR Kit is convenient system for setting up one-tube RT-PCR reactions. It contains Master Enzyme Mix including highly processive dART reverse transcriptase, "hot start" DNA polymerase and unique RNase Inhibitor working well at elevated temperature. Master Buffer contains optimized 2 x buffer including dNTPs, stabilizers and reaction enhancements.

Kit is designed for sufficient amplification DNA from any RNA with high specificity and sensitivity in a one-step process. Our system is dedicated for analytic as well as cloning purposes.

| COMPONENT:            | E0803-01 | E0803-02   |
|-----------------------|----------|------------|
| 2 x Master Buffer Mix | 350 µl   | 2 x 0.7 ml |
| Master Enzyme Mix     | 25 µl    | 100 µl     |
| Nuclease-free Water   | 1 ml     | 4 x 1 ml   |

Reagents are provided for 25 or 100 RT-PCR reactions of 25 µl each.

### Protocol:

- In 0.2 ml PCR tube, combine as follows:

| Component:            | Amount:  |
|-----------------------|----------|
| 2 x Master Buffer Mix | 12.5 µl  |
| Sens primer 10 µM     | 1 µl     |
| Reverse primer 10 µM  | 1 µl     |
| RNA (10 ng–2 µg)      | x µl     |
| Master Enzyme Mix     | 1 µl     |
| Nuclease-free Water   | to 25 µl |

- Gently mix reaction by pipetting or if needed briefly centrifuge.
- Transfer the sample to thermal cycler. Incubate as follows: 30 min at 50°C followed by standard PCR with annealing temperatures suitable for the primers.

| Step             | Temperature | Time       | Number of Cycles |
|------------------|-------------|------------|------------------|
| Pre-denaturation | 94°C        | 5 min      | 1                |
| Denaturation     | 94°C        | 30 s       | 30-40 cycles     |
| Annealing        | 50-65°C     | 30 s       |                  |
| Extension        | 72°C        | 1 min/1kb  |                  |
| Final Extension  | 72°C        | 5 min      |                  |
| Cooling          | 4°C         | Indefinite | 1                |

- Analyze 5-20 µl of RT-PCR sample by agarose gel electrophoresis with suitable molecular markers.

This product is developed, designed and sold exclusively for research purposes and in vitro use only.

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