# 5x ZipTaq PCR Kit

Cat #: 324



Amount: 125 ul enyzme

 $2 \times 1.25$  ml tubes of 5X ZT-PCR Mix (sufficient for 500 x 25  $\mu$ l reactions)

**Shipping conditions:** Ambient

**Storage conditions:** For best performance store at -20°C

**Thermostability:** Retains at least 85% activity after 1 hour at 95°C

**Shelf life:** At least 1 year if stored at -20°C and 10 freeze/thaws or at least 1 month if stored at 4°C.

Expiration: On tube label

### PRODUCT DESCRIPTION:

Our PCR kit contains 5X concentrated master mix (5X ZT-PCR Mix) lacking only the ZipTaq enzyme. The enzyme is provided in a separate vial, which allows an adjustment of its final concentration in PCR.

ZipTaq DNA Polymerase is a Taq polymerase with multiple mutations that make it the fastest enzyme in our collection. With the addition of one of our PECs, reactions can tolerate up to 40% blood. This kit can be used for conventional as well as real-time PCR. For real-time applications you may need to add a fluorescent dye as an alternative to probes. Concentration of dNTPs is 1mM each.

#### TYPICAL PCR PROTOCOL for a 25 µl reaction:

Reagent	Volume	Final Concentration
5x ZT-PCR Mix	5 μl	1x
Left Primer	variable	200 nM
Right Primer	variable	200 nM
DNA template†	variable	0.1-100 ng
PCR Enhancer Cocktail (recommended)*		
ZipTaq	0.05 – 0.25 μl **	
De-ionized distilled H2O	Adjust final volume to 25 µl	

<sup>†</sup> DNA amount depends mostly on genome size and target gene copy number.

## 3-STEP CYCLING CONDITIONS (For 25 ul reactions):

- 1. Initial Denature: 95° for 1 minutes for 1 cycle \*
- 2. Denaturing: 94° for 1-5 seconds †
- 3. Annealing: 50°-68° depending on the specific Tm primers for 1-5 seconds †
- 4. Extension: 68° for 1-5 seconds/kb target †
- 5. Repeat steps 2-4 for 25-40 cycles

## 2-STEP CYCLING CONDITIONS (For 25 ul reactions):

- Initial Denature: 95° for 1 minutes for 1 cycle \*
  Denaturing: 94° for 1-5 seconds †
- 3. Annealing/Extension: 60°-65° depending on the specific Tm primers for 1-5 seconds/kb target †
- 4. Repeat steps 2 and 3 for 25-40 cycles

<sup>\*</sup> For optimal performance, we recommend using one of our PCR Enhancer Cocktails (PEC-1, PEC-1GC, PEC-2, or PEC-2-GC) or 1.3 M Betaine, a general PCR enhancer.

<sup>\*\*</sup> To determine specific optimal enzyme concentration, we strongly recommend an enzyme titration test for each target. A good starting amount of the enzyme per 25  $\mu$ l reaction is 0.05  $\mu$ l for purified DNA templates and 0.25  $\mu$ l for crude samples containing 5-10% whole blood, plasma or serum. Targets larger than 1 kb may require more enzyme or may benefit from the use of an LA (Long Accurate) version of the polymerase.

<sup>\*</sup> A 2-5 minute initial denaturation is recommended for crude samples containing 5-10% whole blood, plasma or serum.

<sup>†</sup> Exact number of seconds will depend on the thermocycler and target. We recommend experimentation to determine precise cycling parameters.