

# Truin Science Ltd. Ready Taq (1 ml)

Cat. # ETS4013

### Contents:

2x Tag Mix 1 ml

Nuclease-free water 1 ml

\*Store at -20°C

\*For research purposes only.

Recommendations for Template DNA in 50µl reaction volume.

Human genomic DNA	0.1-1 μg
Plasmid DNA	0.5-5 ng
Phage DNA	0.1-10 ng
E.coli genomic DNA	10-100 ng

- 2. Gently vortex the sample and briefly centrifuge to collect all drops to the bottom of the tube.
- 3. Overlay the sample with mineral oil or add an appropriate amount of wax.
- \*This step may be omitted if the thermal cycler is equipped with a heated lid.

# Description

2x Taq mix is a premixed, ready to use solution containing Taq DNA Polymerase, dNTPs, Mg<sup>2+</sup>, Reaction Buffer and Loading Buffer at optimal concentrations for efficient amplification of DNA templates by PCR. To prepare the final PCR, only primers and template DNA are to be added. This premixed formulation saves time and reduces contamination due to the fewer pipetting steps required for PCR set up. The mix retains all features of Taq Polymerase.

Taq DNA Polymerase is a thermostable recombinant DNA polymerase derived from thermophilic bacterium *Thermus aquaticus*. Its molecular weight is 94 kDa. Taq DNA Polymerase can amplify DNA target up to 5 kb. The elongation velocity is 0.8~1.0kb/min (70~75°C). It has 5′ to 3′ polymerase activity but lacks 3′ to 5′ exonuclease activity. It generates 3′-dA overhangs in PCR reactions.

4. Perform PCR using the following thermal cycling conditions.

Initial Denaturation	94°C	3 minutes
25 -35 Cycles	94°C 50-68°C* 72°C	30 seconds 30 seconds 1 minute**
Final Extension	72°C	10 minutes

\*Annealing temperature depends on the particular primer sequences used.

Note: as a result of the composition of the ReadyTaq mix, annealing temperatures may vary slightly from those optimized under standard conditions.

\*\* ~1 min per kb of product being amplified.

Note: as a result of the composition of the ReadyTaq mix, required extension times may be slightly longer than what has been optimized under standard conditions.

# **Applications**

- High throughput PCR.
- Routine PCR with high reproducibility.
- Generation of PCR products for TA cloning.

#### **Product use limitation**

This product is developed, designed and sold exclusively for research purposes and in vitro use only. The product was not tested for use in diagnostics or for drug development, nor is it suitable for administration to humans or animals.

#### **Feature**

- Convenient Taq DNA Polymerase in a ready-to-use Mix.
- High yields of PCR products with minimal optimization.
- Reproducible lower contamination and pipetting error risk.
- Set up PCR mixtures in a laminar flow cabinet equipped with a UV lamp.
- Wear fresh gloves for DNA purification and reaction set up.
- Use reagent containers dedicated for PCR. Use positive displacement pipettes, or use pipette tips with aerosol filters to prepare DNA samples and perform PCR set up.
- Always perform "no template control" (NTC) reactions to check for contamination.

## **Quality Control**

The absence of endodeoxyribonucleases, exodeoxyribonucleases and ribonucleases is confirmed by following quality tests. Activity tested by amplification of a single-copy gene from human genomic DNA.

#### **Endodeoxyribonuclease Assay**

No detectable conversion of covalently closed circular DNA to a nicked DNA was

# Composition of the Taq Mix

Taq DNA polymerase is supplied in 2x Taq buffer, dNTPs, 3 mM MgSO<sub>4</sub> and bromophenol blue. Taq mix buffer is a proprietary formulation optimized for robust performance in PCR.

#### **Protocol**

nuclease free

All solutions should be thawed on ice, mixed completely and briefly centrifuged.

# 1. Add the following to a PCR tube on ice: For a total 50µl reaction volume.

Component Volume Final of sample concentration 25 µl Tag Mix (2x) 1x Forward variable  $0.1-1 \, \mu M$ Primer 0.1-1 μM Reverse variable Primer Template variable 10 pg-1 μg DNA Water To 50 ul

Observed after incubation of 25 $\mu$ l Taq Mix (2x) with 1 $\mu$ g of pBR322 DNA in 50 $\mu$ l for 4 hours at 37°C and at 70°C.

#### **Exodeoxyribonuclease Assay**

No detectable degradation of lambda DNA-HindIII fragments was observed after incubation of  $25\mu l$  of Taq Mix (2x) with  $1\mu g$  of digested DNA in  $50\mu l$  for 4 hours at  $37^{\circ}C$  and at  $70^{\circ}C$ .

#### **Ribonuclease Assay**

0% of the total radioactivity was released into trichloroacetic acid-soluble fraction after incubation of 25 $\mu$ l of Taq Mix (2x) with 1 $\mu$ g of E. coli [3H]-RNA (40000cpm/ $\mu$ g) in 50 $\mu$ l for 4 hours at 37°C.

0% of the total radioactivity was released into trichloroacetic acid-soluble fraction after incubation of 25 $\mu$ l of Taq Mix (2x) with 1 $\mu$ g of E. coli [3H]-RNA (40000cpm/ $\mu$ g) in 50 $\mu$ l for 4 hours at 70°C.