



## E.Z.N.A.® Direct Tissue PCR Kit

Product TQ2300

### Introduction

The E.Z.N.A.™ Direct PCR kits are an innovative buffer systems that radically simplifies the extraction of nucleic acids from a variety of sources. These kit contents reagents that quickly release the genomic DNA from samples and neutralize the inhibitors. The lysate is ready for PCR amplification without any further purification procedure.

### Overview

The E.Z.N.A.™ Tissue Direct PCR Kit provides an easy and rapid method for the extraction of genomic DNA for consistent PCR analysis. Animal tissue sample Include moused tail, ear punch, and cultured cells is first lysed in lysis Buffer (L1 Buffer) in the presence of Proteinase K. After a short heating incubation to inactivate the proteinase, the lysate is neutralized with PCR Neutralization buffer. The lysate is then ready for PCR amplification.

Product Number	TQ2310-00	TQ2310-01	TQ2310-02
Purification	20 Preps	100 Preps	500 Preps
L1 Buffer	2.5 ml	12 ml	60 ml
L2 Buffer	1 ml	4 ml	20 ml
PCR Neutralization Buffer	5 ml	25 ml	125 ml
Proteinase K	10 mg	50 mg	250 mg
Direct PCR Buffer	10 ml	50 ml	250 ml
Instruction Manual	1	1	1

### Storage and Stability

All components of the Tissue Direct PCR Kit are stable for at least 12 months from the date of purchase when stored at 22°C-25°C. After dissolved in L2 Buffer, Proteinase K should be stored at -20 C. For long term storage.

## Material to Be Provided by User

- Water bath or heating block at 56 °C
- Water bath or heating block at 95 °C
- Sterile 1.5 ml microfuge tubes
- Tabletop microcentrifuge capable of 10,000 x g

## Before starting:

Please take a few minutes to read this booklet thoroughly and become familiar with the protocol. Prepare all materials required before starting.

- Prepare protein Proteinase K solution (20 mg/ml) as following:

TQ2300-00	Add 500 µl L2 Buffer into each vial
TQ2300-01	Add 2.5 ml L2 Buffer into each vial
TQ2300-02	Add 12.5 ml L2 Buffer into each vial

- Please read the entire booklet to become familiar with the E.Z.N.A.™ Direct Blood PCR procedure.
- Choose the most appropriate protocol to follow. Protocols are described for each of samples.
- Follow general lab protection procedures such as wearing gloves, safety glass when handling any reagent supplied with the kit. Avoid contact skin.

## Tissue Direct PCR Protocol

1. Cut 5-10 mg animal, ear punch or mouse tail ampute and place into a 1.5 ml microcentrifuge tube.
2. Add 100 µl Buffer L1 Buffer and 20 µl Proteinase K solution (dissolved in Buffer L2 Buffer) and mix thoroughly by pipetting or vortexing.
3. Incubate at room temperature for 56 °C for 10 minutes.
4. Incubate at 95 °C for 3 minutes. The tissue will not be completely digested at end of the incubation. It is normal and will not effect performance.
5. Add 100 µl PCR Neutralization Buffer to the lysate. Mix throughly by vortexing. Centrifuge at 13,000 x g for 5 minutes at room temperature.
6. Transfer the supernatant to a new 1.5 ml centrifuge tube and store at 4 °C or use immediately in PCR reaction.
7. PCR amplification: Add 1-5 µl lysate to 20-50 µl reaction using Direct PCR Buffer (supplied). Add primers, Taq polymerase, dNTP and MgCl<sub>2</sub> for PCR amplification. **Note:** the volume of lysate should not exceed 10% of total PCR reaction volume.