





Introduction

The **DNrich** Tissue Kit provides all of the reagents necessary to extract DNA from a wide variety of biological sources. DNA purified with this kit is suitable for a variety of applications, including amplification and digestion with restriction endonucleases.

DNrich Tissue Kit components*

Cat. No. AEDX-1002-050

TD Buffer	20 ml
Activator Reagent**	2 ml
VI Buffer	15 ml
PE Buffer	15 ml
Wash Buffer (conc.) **	15 ml
Elution Buffer	3 ml
Manual	1

^{* 1.5} ml microcentrifuge tube, molecular biology grade water, and Absolut ethanol are needed but are not included.

Chemical Hazard

Always wear gloves and practice standard safety precautions while using the kit. Do NOT disinfect extraction waste in solutions containing **bleach** or any other form of acid. To clean any items contaminated with the reagent, simply soak in detergent and water to remove all traces of contamination before cleaning with bleach or acidic solutions.

Reminder

Pre-set heather block at 65°C.

Prepare Activator Reagent immediately prior to use. Prepared Activator Reagent must be kept at 4°C.

Activator Reagent Preparation

Add 2 ml of molecular biology grade water to **Activator Reagent**, and vortex it well.

Note: Mark the check box on the bottle and write the date.

Note: For the best results, the prepared Activator Reagent should be used immediately. Prepared Activator Reagent can be stored for up to 3 months or 6 months at 4°C and -20°C, respectively.

Wash Buffer Preparation

Add 35 ml molecular biology grade Absolute Ethanol to Wash Buffer bottle before first use and mark the check box on it.

^{**} Please refer to Activator Reagent Preparation and Wash Buffer Preparation before using this kit.

PROTOCOL

Step 1: Sample Preparation

• Cut off **50 mg** (up to 200 mg) of tissue and transfer it to a 1.5 ml microcentrifuge tube.

Step 2: **Tissue Digestion**

- Add 400 μl of TD Buffer and 40 μl of prepared activator reagent to the sample tube and vortex vigorously.
- Incubate at 65°C until tissue completely lysed (usually 20 to 120 minutes). **Note:** During incubation time, vortex the sample tube every 10 minutes.
- Incubate at 85°C for 10 minutes.

Step 3: Lysis

- Add 300 µl of VI Buffer to the sample tube and mix by vortex.
- Keep at **room temperature** for **10** minutes and **invert** regularly.
- Centrifuge at **12000** g for **15** minutes.
- Carefully transfer about 300µl of supernatant to a new 1.5 microcentrifuge tube and do not disturb the phases.

Step 4: **DNA Precipitation**

- Add 300 μl of PE Buffer to the sample tube and mix by invert for 15 times.

 Optional: Incubate the samples at -20°C for 30 to 60 minutes. In sample with low amount of DNA, incubation time can be up to overnight.
- Centrifuge at 12000 g for 15 minutes.
- **Decant** the supernatant into waste. **Note**: While decanting, be careful not to lose the DNA pellet.

Step 5: Wash

- Add 500 μl of Wash Buffer to the sample tube, invert for 10 times and then centrifuge at 12000 g for 10 minutes.
- **Decant** the supernatant into waste.
- Add 500 μl of Wash Buffer to the sample tube, invert for 10 times and then centrifuge at 12000 g for 10 minutes.
- **Decant** the supernatant into waste, and let stand until dry or incubate at 55°C for 10 minutes.

Step 6: **DNA Elution**

- Add **50 µl of Elution Buffer** to the sample tube.
- Resuspend the DNA for 5 minutes at 55°C, and then centrifuge at 12000 g for 1 minute. Note: Store DNA at -20°C.

