



Introduction

The **DNrich Plant Kit** is designed for DNA extraction from different sources of plants such as leaf or seed. The quality of extracted DNA can be varied, depending on the samples used. The kit is intended for downstream application of molecular biology.

DNrich Plant Kit components*

Cat.	No.	AEPDX.	-1004-050
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A solution**	25 ml
B Solution	15 ml
C Solution	15 ml
Wash Solution (conc.) *	15 ml
Elution Buffer	3 ml
Manual	1

^{* 1.5} ml micro-centrifuge tube, molecular biology grade water, ethanol absolute and 70% ethanol are needed but are not included.

Caution

The components contain irritants. During operation, always wear a lab coat, disposable gloves, and protective goggles.

Reminder

Pre-set heather block at 56°C.

Prepare A Solution immediately prior to use.

Pre-warmed A Solution and Elution buffer at 56°C.

A Solution preparation

Add 22 ml of molecular biology grade water to A Solution, and shake it well.

Note: mark the check box on the bottle. Note: With **1 M NaOH** adjust the pH to 8.

Note: For the best results, the prepared A Solution should be used immediately. Prepared A Solution can

be stored for up to one month at 4°C.

Wash Solution 1 & 2 Preparation

Add **35 ml** molecular biology grade **absolute ethanol** to each **Wash Solution** before first use and check mark on the bottle.

^{**} Please refer to A Solution Preparation and Wash Solution before using this kit.

PROTOCOL

Step 1: Sample Preparation

- Cut off **50 mg** (up to 200 mg) of plant tissue.
- **Grind** the sample to a fine powder and transfer it to a 1.5 ml micro-centrifuge tube. Note: Some plant samples need liquid nitrogen and/or washed and autoclaved sand to be ground sufficiently.

Step 2: Lysis

Note: Prepare A Solution immediately prior to use.

- Add 500 µl of pre-warmed A Solution into the sample tube and mix by vortex.
- **Incubate at 56°C** for 90 minutes. During incubation, vortex vigorously the tube every 30 minutes.

Note: Incubation time could be varied from 30 minutes to overnight for different plant samples.

- Add 300 µl of B Solution and mix by vortex for 30 seconds.
- Keep at room temperature for 10 minutes, and invert every 5 minutes.
- Centrifuge for 15 minutes at **11500** g.
- Carefully transfer about $300\mu l$ of supernatant into a new 1.5 micro-centrifuge tube and do not disturb the phases.

Note: The amount of supernatant phase varies.

Step 3: **DNA Precipitation**

- Add 300 µl of C Solution to sample tube and mix by invert for 15 times.
- Incubate the samples at -20°C for 30 minutes.
- Centrifuge at **12000** g for 10 minutes.
- **Decant** the supernatant into waste.

Note: While decanting, be careful not to lose the DNA pellet.

Step 4: Wash

- Add 500 μl of Wash Solution 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 Solution 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.

Step 5: **DNA Elution**

- Add $50 \mu l$ of Elution buffer to the sample tube.
- Allow to re-suspend the DNA for 5 minutes at 55°C, and then centrifuge at **8000 g** for 1 minute. Note: Store DNA at **-20**°C.

