**Cat. No. AFPRX-1151**

For Research Use Only

# Fast RNrich Plant Kit

**Azma Elixir Pajooh**

## Introduction

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

| **Fast RNrich Plant Kit components *** | **Cat. No. AFPRX-1151-050** |
|---|---|
| **RN Buffer **** | **25 ml** |
| **Beta Activator** | **1 ml** |
| **VI Buffer** | **15 ml** |
| **SE Buffer** | **15 ml** |
| **Wash Buffer(conc.) **** | **15 ml** |
| **Elution Buffer** | **2.5 ml** |
| **column** | **50** |
| **Manual** | **1** |

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

** Please refer to reminder, **RN Buffer** and **Wash Buffer Preparation** before using this kit.

## 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 56°C.

**During RNA extraction, never open the microtube cap outside the laminar hood.**

## RN Buffer preparation

Add **22** ml of molecular biology grade water to **RN Buffer**, 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 RN Buffer should be used immediately. Prepared RN Buffer can be stored for up to three months at 4°C.**

## 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.

## PROTOCOL

Step a: **Sample Preparation**

a1 Cut off **50 mg** (up to 200 mg) of plant tissue.
a2 **Grind** the sample to a fine powder and transfer it to a 1.5 ml micro-centrifuge tube.
**Note**: Many plant samples need **liquid nitrogen** and/or washed and autoclaved sand to be ground sufficiently.

Step b: **Tissue Digestion & Lysis**

b1 Add **500 µl** of **RN Buffer** to the sample tube and vortex vigorously.
**Optional**: Add **7 µl** of **Beta Activator** to the sample tube and vortex vigorously
b2 Incubate at **56°C** until tissue are lysed, completely (usually **30 to 120** minutes**).**
**Note**: During incubation time, vortex the sample tube every 10 minutes.
**Optional (to have a higher yield)**

> Add **300 µl of VI Buffer** to the sample tube and mix by vortex for 3 seconds.
> Keep at **RT** for **5 minutes**.

b3 **Centrifuge** at **10000 g** for **5** minutes.
**Note**: Do not disturb the phases.
b4 Carefully transfer about **300µl of supernatant** to a new 1.5 ml tube.
b5 Add **300µl** of SE Buffer to the tube, invert for 5 times and keep at room temperature for **3** minutes and then transfer all the sample into a **spin column**.
b6 Centrifuge at **2000 g** for **2** minutes and **discard** the flow through.

Step c**: Washing**

**Note**: Prepare **Wash Buffer** before first use
c1 Add **500 µl** of **Wash Buffer** to the column, centrifuge at **8000 g** for **2** minutes.
c2 Discard the flow through.
c3 **Repeat step c1 and c2.**

Step d: **Column Drying**

d1 Centrifuge at **8000 g** for **1** minute.
d2 **Discard** the flow through and place the column into a new **1.5 ml** microcentrifuge tube.

Step e: **RNA Elution**

e1 Add **40 µl** of **Elution Buffer** to the center of column and let stay at **RT** for **3** minutes.
e2 Centrifuge at **10000 g** for **2** minutes.
**Note**: Eluted RNA is ready for downstream analysis and should be stored at **-20°C**.