## NAUTIAGENE

**Product Information** 

# NautiaZ Plant Total RNA Mini Kit

#### (100/300 prep)

Cat.No.: NGPRZ-S100/NGPRZ-S300 Sample: up to 100 mg of Tissue

up to 50 mg of dry plant Tissue

Yield: Up to 30 μg

NGCRZ-S100	NautiaZ Cell/Blood Total RNA Mini Kit (100 prep)
NGCRZ-S300	NautiaZ Cell/Blood Total RNA Mini Kit (300 prep)
NGBRZ-S100	NautiaZ Bacteria/Fungi RNA Mini Kit (100 prep)
NGBRZ-S300	NautiaZ Bacteria/Fungi RNA Mini Kit (300 prep)
NGTRZ-S100	NautiaZ Tissue Total RNA Mini Kit (100 prep)
NGTRZ-S300	NautiaZ Tissue Total RNA Mini Kit (300 prep)
NGPRZ-S100	NautiaZ Plant Total RNA Mini Kit (100 prep)
NGPRZ-S300	NautiaZ Plant Total RNA Mini Kit (300 prep)
NGMRZ-S050	NautiaZ microRNA Mini Kit (50 prep)
NGVN-S100	Nautia Viral Nucleic Acid Extraction Kit (100 prep)
NGVN-S300	Nautia Viral Nucleic Acid Extraction Kit (300 prep)

#### **Contents**

	NGPRZ-S100T	NGPRZ-S100	NGPRZ-S300
PR Buffer	2 ml x2	110 ml	105 ml x3
W1 Buffer	2 ml	45 ml	125 ml
W2 Buffer*	300 ul x2	15 ml	25 ml x2
Elution Buffer	1 ml	10 ml	30 ml
RZ Column	4 pcs	100 pcs	300 pcs
Collection Tube	4 pcs	100 pcs	300 pcs
User Manual	1	1	1

<sup>\*</sup>Add 1.2 ml x2 / 60 ml / 100 ml x2 ethanol (96-100%) to W2 Buffer prior to the initial use

### **Buffer Preparation**

• Add ethanol (96-100%) to the Wash Solution prior to first use:

	NGPRZ-S100T	NGPRZ-S100	NGPRZ-S300
W2 Buffer	300 ul x2	15 ml	25 ml x2
ethanol (96 ~ 100%)	1.2 ml x2	60 ml	100 ml x2

### Additional Requirements

- 1. ß Mercaptoethanol
- 2. RNase-free microcentrifuge tubes
- 3. isopropanol
- 4. ethanol

#### Important Notes

- 1. Buffer contains chaotropic salt is harmful and irritant agent.
- 2. Use sterile, RNase-free pipet tips and microcentrifuge tube. Wear a lab coat and disposable gloves to prevent RNase contamination.
- 3. Make sure the starting sample amount is under the limit.
- 4. Add ethanol (96-100%) to W2 Buffer prior to the initial use
- 5. All purification steps should be carried out at room temperature.
- 6. All centrifugations should be carried out in a table-top microcentrifuge at  $>12000 \times g$  (10,000-14,000 rpm, depending on the rotor type).

#### **Quality Control**

The quality of NautiaZ Plant Total RNA Mini Kit is tested on a lot-to-lot basis. The kits are tested by isolation of total RNA from plant tissue. More than 1  $\mu$ g of total RNA was quantified with a spectrophotometer and checked by formaldhyde agarose gel analysis. Finally, RT-PCR was used to ensure the quality of total RNA.

#### Storage

Store at room temperature.



Cut off Plant sample Grind the sample under liquid nitrogen to a fine powder.

Add 1 ml PR Buffer
Add 10 μl β-mercaptoethanol
Incubate at 70 °C for 30 mins
until it is completely dissolved.

centrifuge and transfer supernatant to a new 1.5 ml tube

Add 1/2 volume isopropanol
Shake or pipetting.



Centrifuge for 0.5 min
Discard the flow-through

Add 400 µl W1 Buffer Centrifuge for 30 sec Discard the flow-through

Add 600 µl W2 Buffer Centrifuge for 30 sec Discard the flow-through



Add 50-200 μl Elution Buffer Stand at RT for 2 mins

Centrifuge for 2 mins to elution RNA





**Cell Lysis** 















**Elution** 

### PURIFICATION PROTOCOLS

STEP	PROCEDURE
1 Sample prepare	Cut off up to 100 mg fresh plant tissue or up to 50 mg dry plant tissue. Grind the sample under liquid nitrogen to a fine powder by using a mortar and pestle.
2 Cell Lysis	Add 1 ml PR Buffer and 10 $\mu$ l of $\beta$ - Mercaptoethanol to the sample in the mortar and grind the sample until it is completely dissolved.
	Transfer the sample mixture to a RNase-free microcentrifuge tube and incubate at 70°C for 30 mins.  Invert the tube every 10 mins.  Centrifuge at 2-8°C at 14,000 x g for 10 mins and transfer the supernatant to a new microcentrifuge tube.
	Add 1/2 volume of isopropanol to the sample and shake vigorously. Place a RZ column in Collection Tube.
3 RNA Binding	Transfer all of the sample mixture (up to 700 $\mu$ l once) to RZ column and centrifuge 30 senconds at 14,000 x g. Discard the flow-through and place RZ Column back in the Collection tube.
4-1 Wash	Add 400 $\mu$ l W1 Buffer to RZ Column. Centrifuge at 14,000 x g for 30 seconds. Discard the flow-through and place RZ Column back in the Collection tube.

STEP	PROCEDURE
Optional Step: DNase	If DNA-free RNA is required, perform this optional step.  Add 150 µl W2 Buffer (ethanol added) into the RZ column.  Centrifuge at full speed (16,000 x g) for 30 seconds.  Discard the flow-through and place the RZ Column back in the Collection Tube.
	For each isolation reaction , premix <b>80 µl DNase I</b> Incubation Buffer with <b>2 µl DNase I</b> in a new sterile tube (Do not vortex!). Add <b>82 µl of the DNase I solution</b> into the center of the RZ Column membrane and incubate at room temperature for 15 min.
4-2 Wash	Add 600 μl W2 Buffer (ethanol added) to RZ Column. Centrifuge at 14,000 x g for 30 seconds. Discard the flow-through and place RZ Column back in the Collection tube.
5 Dry	Centrifuge at 14,000 x g for 2 minutes to dry the column.
6 Elution	Place RZ Column to a clean 1.5 ml microcentrifuge tube (not provided).  Add 50-200 µl of Elution Buffer into the center of the column matrix.
	Stand at room temperature for 2 minutes. Centrifuge at 14,000 x g for 2 minutes to elute purified RNA.

STEP	PROCEDURE
7 Pure RNA	Store the RNA fragment at -80 °C.