

EcoSpin Plant Total RNA Kit

50 rxns

Cat No: E2096

Shipping : Ship at ambient temperature.

Storage : Store the kit between 15°C and 25°C.

Store *EcoSpin* Lysis/Binding Buffer at 4-8°C upon receipt.

General Information

EcoSpin Plant Total RNA Kit is designed as a simple and convenient purification of high-quality total RNA from up to 300 mg of plant material. This kit utilizes chaotropic ions and silica-based membrane technology, eliminating the need for expensive resins, β -mercaptoethanol, or time-consuming alcohol precipitation. The standard protocol lasts less than 40 minutes at room temperature and purified RNA can be effectively used in routine downstream applications including cDNA synthesis, northern blotting, differential display, primer extension, and mRNA selection.

Kit Contents

<i>EcoSpin</i> Lysis/Binding Buffer*	(28 ml)
<i>EcoSpin</i> Wash Buffer 1	(22 ml)
<i>EcoSpin</i> Wash Buffer 2**	10 ml concentrate)
<i>EcoSpin</i> Elution Buffer	(5 ml)
<i>EcoSpin</i> Columns	(50)
<i>EcoSpin</i> Collection Tubes	(50)

*Keep *EcoSpin* Lysis/Binding Buffer at 4-8°C upon receipt.

**Add 40 ml absolute ethanol

Protocol for Plant Total RNA

Each isolation procedure is suitable for isolation of total RNA from up to 300 mg of plant material.

1a. Cut the plant tissue sample and determine the amount of tissue by weighing. Grind the sample in a mortar that contains an appropriate amount of liquid nitrogen to cover the sample. Grind the plant tissue thoroughly using a pestle. Allow the liquid nitrogen to evaporate, without allowing the plant tissue to thaw. Immediately, add 500 μ l *EcoSpin* Lysis/Binding Buffer to grinded tissue. Transfer the grinded and homogenized tissue sample into an RNase-free 1.5 ml microcentrifuge tube (not provided). Mix well by vortexing for 10-15 seconds.

1b. Cut the tissue sample and determine the amount of tissue by weighing. Transfer the tissue sample into an RNase-free 1.5 ml microcentrifuge tube (not provided). Add 500 μ l *EcoSpin* Lysis/Binding Buffer. Immediately and vigorously homogenize using a conventional rotor-stator homogenizer with a stainless-steel probe at 15,000 rpm for 30 seconds.

2. Incubate samples at 60°C for 5 minutes to enhance the lysis.

Note: Tightly close the caps of the microcentrifuge tube to prevent evaporation of the *EcoSpin* Lysis/Binding Buffer

3. Add 100 μ l chloroform to the lysate and mix well by shaking. Incubate for 3 minutes at room temperature.

4. Centrifuge the tube at 12000 rpm for 15 minutes at 4°C.

5. Carefully transfer the upper phase to a new RNase-free 1.5 ml microcentrifuge tube (not provided). It is important to not contaminate the upper phase with the lower yellow phases.

6. Add equal volume of 70% ethanol and mix well by shaking.

7. Insert an *EcoSpin* Column into a Collection Tube and transfer 700 μ l sample from step 6 to the *EcoSpin* Columns. Centrifuge at maximum speed in a tabletop microcentrifuge 30 seconds at room temperature. Depending on your lysate volume, repeat Step 7 as necessary.

8. Discard the flow through and add 400 μ l *EcoSpin* Wash Buffer 1 to the *EcoSpin* Column. Centrifuge at maximum speed in a tabletop microcentrifuge 30 seconds at room temperature.

9. Discard the flow through and add 500 μ l *EcoSpin* Wash Buffer 2 to the *EcoSpin* Column. Centrifuge at maximum speed in a tabletop microcentrifuge 30 seconds at room temperature.

10. Discard the flow through and add 200 μ l *EcoSpin* Wash Buffer 2 to *EcoSpin* Column at maximum speed for 2 minutes to completely remove any residual wash buffer.

11. Transfer the *EcoSpin* Column to a clean RNase-free 1.5 mL microcentrifuge tube (not provided).

12. Add 50-100 µL of *EcoSpin* Elution Buffer to the center of the *EcoSpin* Column membrane and incubate the column at room temperature for 1 minute.

13. Centrifuge at maximum speed in a tabletop microcentrifuge 30 seconds at room temperature. Discard the *EcoSpin* Column and store the purified RNA at -20°C (for a few days) or -80°C (for long term storage) until use.