FavorPrep™ Soil RNA Isolation Mini Kit

Cat.No. : FASRK 004, 4 preps
FASRK 050, 50 Preps
FASRK 100, 100 Preps

(For Research Use Only)

Kit Contents:

	FASRK 004 (4 preps_sample)	FASRK 050 (50 preps)	FASRK 100 (100 preps)
Glass Beads	4 vials	50 vials	100 vials
SR1 Buffer	5 ml	60 ml	120 ml
SR2 Buffer	0.3 ml	4 ml	8 ml
SR3 Buffer	0.6 ml	8 ml	16 ml
SR4 Buffer	1.5 ml	20 ml	40 ml
Wash Buffer (concentrate) *	1.5 ml	15 ml	30 ml
RNase-free water	1.5 ml	20 ml	40 ml
RNA Mini Column	4 pcs	50 pcs	100 pcs
Collection Tube	8 pcs	100 pcs	200 pcs
Elution Tube (1.5 ml tube)	4 pcs	50 pcs	100 pcs
2.0 ml tube	4 pcs	50 pcs	100 pcs
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* Preparation of Wash Buffer by adding ethanol (96% ~100%) for first use:			
Cat. No:	FASRK 004	FASRK 050	FASRK 100
ethanol volume for Wash Buffer	6 ml	60 ml	120 ml

Specification:

Principle: spin column (silica membrane)
Sample: 0.25 ~ 1 g
Operation time: < 60 min
Elution volume: 40 µl

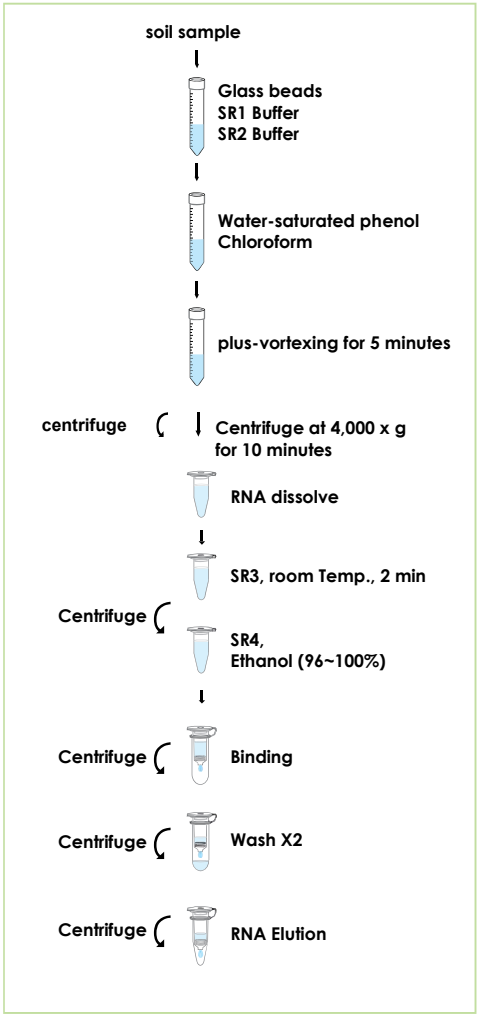
Important Notes:

1. Make sure everything is RNase-free when handling RNA.
2. Buffers provided in this system contain irritants. Wear gloves and lab coat when handling these buffers.
3. Add indicated volume of ethanol (96-100%) to Wash Buffer before use.
4. Phenol and chloroform are hazardous to human health. perform the procedures involving phenol or chloroform in a chemical fume hood.
5. Preheat RNase free water to 60 °C for elution step.

Additional material to be provided by user:

Ethanol (RNase-free, 96~100%)
Isopropanol
Water-saturated phenol
Chloroform
Microcentrifuge for 1.5 ml and 2.0 ml tube capable of at least 13,000 x g
Centrifugefugr for 15 ml tube capable of at least 4,000 X g
Water Bath or Dry Bath
1.5 ml centrifuge tube
15 ml centrifuge tube

Brief Procedure:



General Protocol:

Please Read Important Notes Before Starting Following Steps.

1. Add the Glass Beads (provided) and 1 g of soil sample to a 15 ml centrifuge tube (not provided).
   --If the sample is liquid, add 500 µl of sample to the 15 ml Beads Tube.

2. Add 1 ml of SR1 Buffer to the sample, vortex at maximum speed for 1 minute.

3. Add 50 µl of SR2 Buffer to the sample, vortex at maximum speed for 1 minute.

4. Add 1 ml of water saturated-phenol and 200 µl of chloroform to the sample, mix well by plus-vortexing for 5 minutes to lysis the sample.
   Caution! Phenol and chloroform are hazardous, performing Step 4 and Step 5 in a chemical fume hood.

5. Centrifuge at 4,000 x g for 10 minutes to form aqueous and organic phase.

6. Carefully transfer the upper aqueous phase to a 2.0 ml centrifuge tube (provied). Measure the volume of the upper aqueous.
   -- Carefully transfer the upper aqueous phase and do not disrupt the inter phase that consist of phenol and chlorofrom.
   -- Avoid pipetting any debris and pellet.

7. Add 1 volume of isopropanol, vortex to mix well. centrifuge at full speed (13,000 x g) for 10 min to form a pellet.

8. Carefully discard the supernatant and invert the tube on the paper towel for 5 min to remove residual liquid.
   -- Do not disrupt the pellet when discard the supernatant.
   -- Depending on the soil type, the dark color of pellet is not consist .

9. Add 200 µl of RNase-free ddH2O, vortex to dissolve the pellet completely.
   -- Incubate the tube at 45°C for 10 min if the pellet is hard to dissolve.

10. Add 100 µl of SR3 Buffer to the sample, mix well by vortexing. Incubate the sample at room temperature for 2 minutes.
    -- Note: SR3 Buffer must be suspended completely by vigorously vrotexing before every using.
    -- use 1ml pipettor and cut off the end of 1 ml tip to make it easier for pipetting the SR3 Buffer.

11. Centrifuge at full speed (13,000 x g) for 2 minutes.

12. Carefully transfer the clarified lysate to a 1.5 ml microcentrifuge (not provied).
    --Avoid pipetting any debris and pellet.

13. Add 250 µl of SR4 Buffer and 250 µl of ethanol (RNase-free, 96~100%) to the clarified lysate, mix thoroughly by pulse-vortexing.

14. Place a RNA Mini Column into a Collection Tube and transfer all of the sample mixture to the RNA Column. Centrifuge at full speed (13,000 x g) for 1 min then discard the flow-through. Place the RNA Column to a new Collection Tube.

15. Add 650 µl of Wash Buffer (ethanol added) to RNA Mini Column. Centrifuge at full speed (13,000 x g) for 1 min then discard the flow-through.
    --Make sure that ethanol (RNase-free, 96~100%) has been added into Wash Buffer when first open.

16. Repeat step 15 for one more time.

17. Centrifuge at full speed (13,000 x g) for an additional 3 min to dry the RNA Mini Column.
    --Important step! This step will avoid the residual liquid to inhibit subsequent enzymatic reactions.

18. Place RNA Column into a Elution Tube, Add 40 µl of pre-heated RNase-free ddH_2O to the membrane center of the RNA Mini Column. Stand the RNA Column for 2 min at room temperature.
    --Important step! For effective elution, make sure that the RNase-free ddH_2O is dispensed onto the membrane center and is absorbed completely.

19. Centrifuge at full speed (13,000 x g) for 1 min to elute RNA.