avigene
Molecular Biology Products

AviRex™ Plant RNA

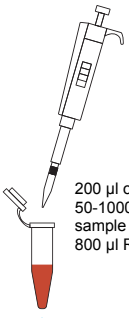
for plant tissue and cell culture

Before Starting

Add 48 ml of absolute ethanol to the PW (only at the first use).

Reagents NOT Provided

1. Chloroform
2. 70% and 96% ethanol

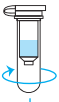


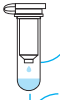
Shake 15 s
RT 8 min

200 µl chloroform
Shake for 15 s
RT 3 min

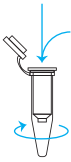
Spin 12 min, 4 ˚C

400 µl of upper phase
Add 400 µl ethanol





Transfer column



Protocol

1. Cutting the tissue into the small pieces on a sterile petri dish by a scalpel and grind with a mortar and pestle under liquid nitrogen. Transfer 50-100 mg of tissue or 6 x 10^6 cells (for cell cultures) into a 1.5 ml tube and add 800 µl of RL solution.
2. Pipetting the tissue into and out of the tip to avoid clumps. You can also homogenize hard tissue by homogenizer on ice. Incubate at room temperature for 8 min.
3. Add 200 µl of chloroform to the mixture. Shake it completely for 15 s and incubate for 3 min at room temperature.
4. Spin for 12 min at 13,000 rpm at 4 ˚C.
5. Transfer 400 µl of the upper phase into a new 1.5 ml tube. Add equal Volume of 70% ethanol (use 96% ethanol for low RNA samples) to the mixture and mix them well.
6. Transfer mixture to the spin column. DO NOT touch upper rim of column. spin for 1 min at 13.000 rpm.
7. Pour off the flow-through of collection tube.
8. Add 700 µl of PW and spin for 1 min at 13.000 rpm.
9. Pour off the flow-through of collection tube. (Optional: repeat step 8 and 9 with 500 µl of PW to have more pure RNA)
10. Spin for 2 min at 13.000 rpm to remove the remaining of the wash buffer . Transfer the spin column to a new 1.5 ml microtube.
11. Add 50 µl of DEPC-treated water, wait 3 min at room temperature. if you want more concentration add less DEPC-treated water (35 µl).
12. Spin for 1 min at 13.000 rpm to elute RNA from the column. Store RNA solution at -70 C.

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kit content

No.	Description:	Packing	Qty.
1	RL Buffer (RNA Lysis Buffer)	20 ml	2
2	PW Buffer (Wash Buffer)	12 ml	1
3	DEPC-treated Water	3 ml	1
4	Spin Column	50	1
5	Collection Tube	50	1

Research Use Only

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