Column extraction of whole blood miRNA kit

Cat: R2200
Package: 25T/50T/100T
Storage: RT, Valid for 1 year.

Product composition:

Kit composition	25T	50T	100T
Lysate A	3mL	6mL	12mL
Lysate B	4.5mL	9mL	18mL
Bleach solution 1	14.85mL	29.7mL	59.4mL
Bleach solution 2	3.75mL	7.5mL	15mL
RNase-free ddH_2O	1mL	2mL	4mL
Adsorption column	25	50	100
2mL collection tube	25	50	100
Specification	1	1	1

Notes:

1. Before use, add β-mercaptoethanol to rinse solution 1 (add 150μL for 25T, 300μL for 50T, and 600μL for 100T). Add absolute ethanol to rinse solution 2 before use. Please refer to the label on the bottle body.
2. Need to own anhydrous ethanol, 95% ethanol and β-mercapto ethanol.

Product description:

This kit uses a unique lysate to rapidly split whole blood (fresh anticoagulant whole blood sample) to separate miRNA and other substances, and then obtains high-purity miRNA through a series of rapid rinse-centrifugation. The extracted miRNA does not contain large RNA and DNA, and can be applied to many downstream experiments such as RT-qPCR. This kit uses one step column, simple operation and short time.

Product features:

1. Completely non-toxic, no need for phenol chloroform and other organic reagents extraction.
2. Wide application range, it can be used for extraction and purification of microRNA and total RNA of a variety of samples.
3. The operation is simple and fast, and the whole process is about 30 minutes.
4. With good purity and high yield, it can be directly used in various downstream experiments.

Operation procedure:

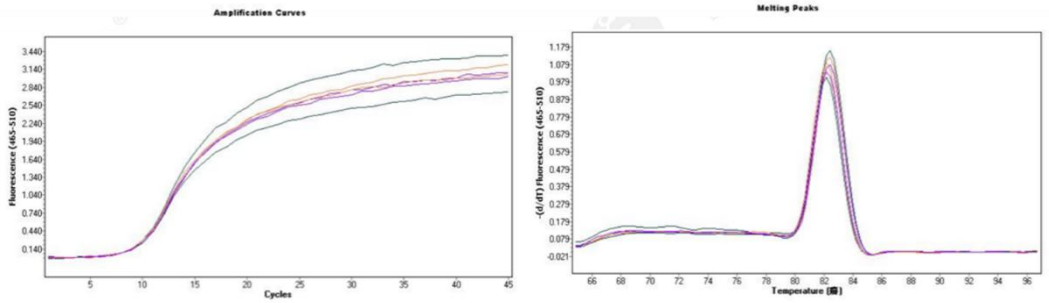
1. Add 100μL lysate A, 150μL lysate B, 4.5μL β-mercaptoethanol and 200μL blood into 1.5mL centrifuge tube successively, swirl for 5s, and let stand at room temperature for 3min.
2. Add 200μL anhydrous ethanol to the above cracked liquid, swirl and shake for 20s, and stand at room temperature for 3min.
3. Centrifuge at 12000rpm for 3min at 4℃.
4. Slowly transfer 400μL supernatant to a new 1.5mL centrifuge tube, add 400μL 95% ethanol and swirl for 5s.
5. Add the above mixture to the adsorption column at one time (the adsorption column is placed in the collection tube), centrifuge at 12000rpm for 1min, and discard the filtrate.
6. Add 300μL bleach solution 1 and centrifuge at 12000rpm for 1min. Discard filtrate.

7. Repeat 6.
8. Add 300μL bleaching solution 2 and centrifuge at 12000rpm for 1min. Discard filtrate.
9. Repeat 8.
10. Put the adsorption column into an empty collection tube, centrifuge it at 12000rpm for 2min, discard the collection tube, and open the adsorption column for 2min to remove the residual ethanol.
11. Put the adsorption column into the new 1.5mL centrifuge tube, central to the adsorption column to join 25μL RNase-free ddH_2O, place 2min at room temperature, 4℃, 12000rpm centrifugal 2min, get the miRNA solution.

Note:

1. All relevant utensil consumables should be RNase-free products. Be careful during operation. Wear masks and gloves to avoid contamination of samples with RNA enzymes in the environment.
2. Avoid volatilization, oxidation and pH value changes caused by long-term exposure to the air, and cover the solution tightly in time after use.
3. Try to use fresh blood for miRNA extraction.
4. The bleaching solution 1 will crystallize out when it is placed at 4℃ for a period of time. At this time, it should be heated and dissolved to room temperature before use.

Example:



The extracted miRNAs can be used to detect specific primers:

The quality of miRNA extracted by this kit was tested, including whole animal blood, serum and plasma. In order to further test whether miRNA extracted by this kit can be specifically applied to fluorescent quantitative PCR, U6 universal primer for mouse blood was designed for verification. Through fluorescence quantitative PCR analysis, it was found that the dissolution curve was relatively neat. It indicates prim-specific amplification. Moreover, the specificity of the melting curve is good and the CT value is close, indicating that this method can obtain the fluorescence quantitative PCR detection of miRNA that can be stably applied to specific primers.

Related products:

R1600 DEPC Treating water
R1050 5×RNA Loading Buffer
M1010 10×MOPS Buffer
R2220 Column extraction of tissue cell miRNA kit
R2230 Column extraction of plant miRNA kit