TIANGEN

(DP316) TIANamp Micro DNA Kit ——Blood

TIANGEN BIOTECH(BEIJING)CO., LTD

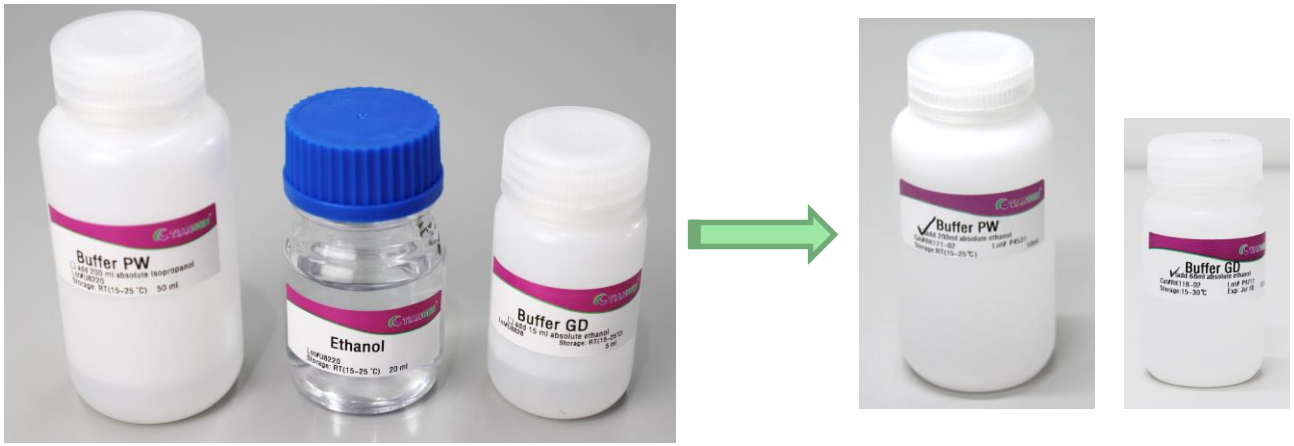
Ver. No. 20170328

Experiment Preparation

1. Anticoagulant blood ≤ 100μl
2. 96-100% ethanol
3. Pipette and matched sterile tips (10 μl, 200 μl, 1 ml); 1.5 ml centrifuge tubes
4. Vortex oscillator; Dry bath/water bath; Centrifuge

Experiment Preparation-Kit Preparation

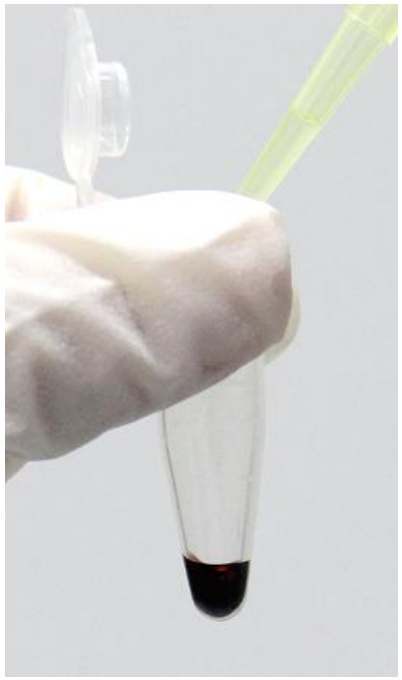
Please add 96-100% ethanol in Buffer PW and GD before use according to the volume indicated on the label of the bottle.



Preparation of Carrier RNA stock solution

When using carrier RNA for the first time, please dissolve Carrier RNA (310 µg) in 310 µl RNase-Free ddH_2O, then aliquot the solution and store at -20℃. The concentration of the solution is 1 µg/µl. Avoid repeated freezing and thawing for more than 3 times.

Step 1



This experiment takes human blood as an example, and the volume of sample is 100 μl.

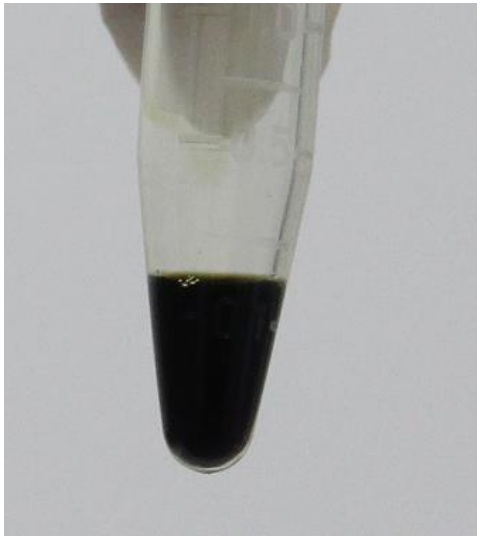
If the sample volume is less than 100 μl, add Buffer GA to 100 μl.

Step 2

Add 10 μl Proteinase K and 100 μl Buffer GB

Mix well upside down. Briefly centrifuge to remove water drops of the cap and inner wall.

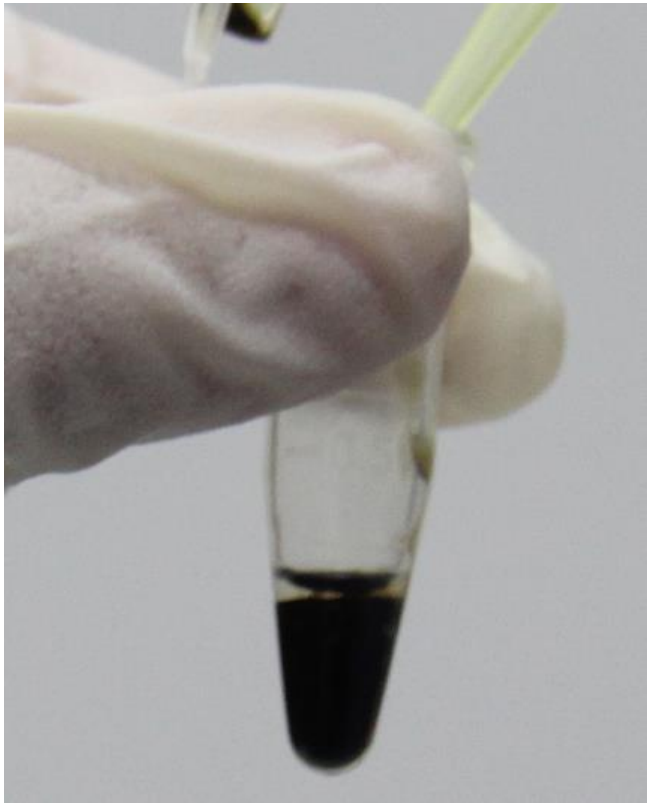
Incubate at 56℃ for 10 min, and mix upside down during the period.



The color will become dark and green after heating, and will become clear without turbidity after shaking.

(If serum/plasma volume <50 μl, add 1 μl of 1μg/μl Carrier RNA storage solution)

Step 3



Add 50 µl ethanol (96-100%). If the room temperature is higher than 25℃, please precool ethanol on ice. Gently mix the sample well upside down, and place at room temperature for 3 minutes. Briefly centrifuge to remove water drops of the cap and inner wall.

Step 4

Transfer the solution from the previous step to Spin Column CR2. (place CR2 into the collection tube)

Centrifuge at 12,000 rpm (~13,400×g) for 30 sec, discard the waste liquid in the collection tube and place the Spin Column CR2 into the collection tube.

Step 5

Add 500 μl Buffer GD to Spin Column CR2 (make sure 96-100% ethanol has been added before use)

Centrifuge at 12,000 rpm (~13,400×g) for 30 sec, discard the waste liquid in the collection tube and place the Spin Column CR2 into the collection tube.

Step 6

Add 600 μl Buffer PW to Spin Column CR2 (make sure 96-100% ethanol has been added before use)

Centrifuge at 12,000 rpm (~13,400×g) for 30 sec, discard the waste liquid in the collection tube and place the Spin Column CR2 into the collection tube.

Step 7 Repeat step 6.

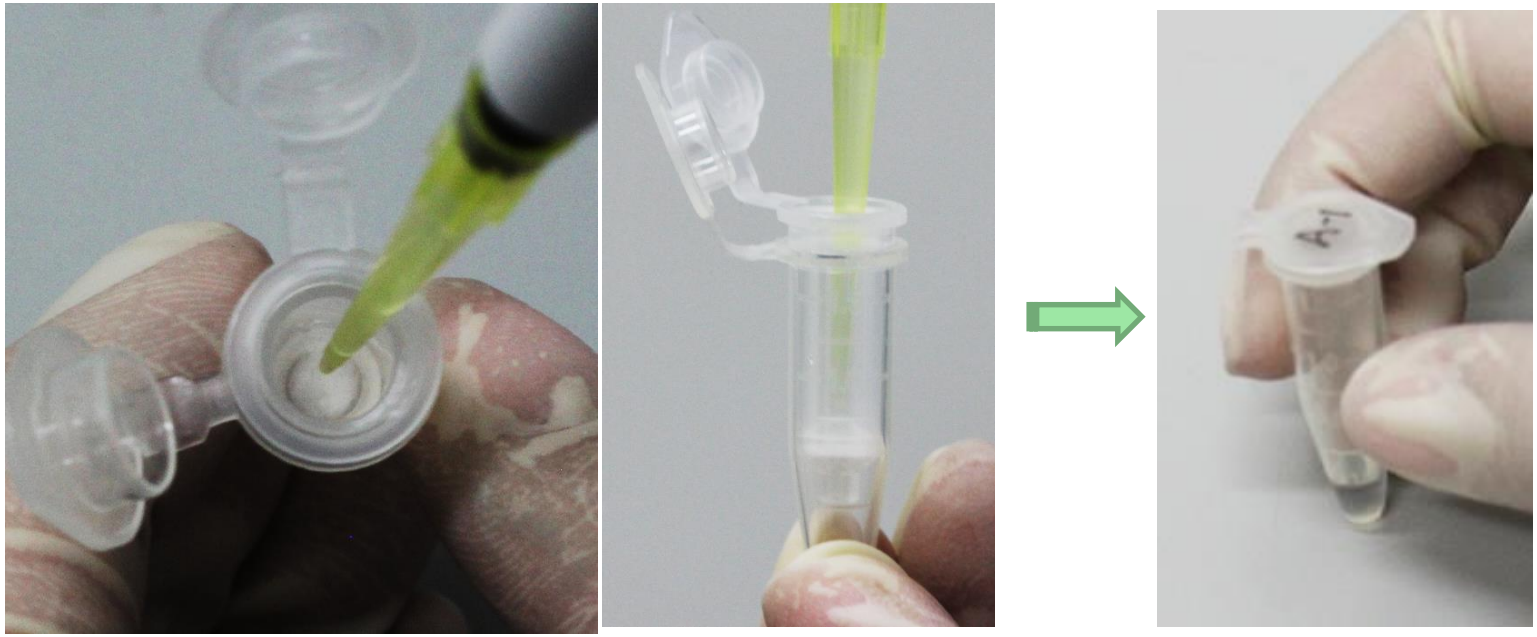
Step 8

Centrifuge at 12,000 rpm (~13,400×g) for 2 min, and discard the waste liquid.

Place the Spin Column CR2 at room temperature for 2-5 minutes to completely dry Buffer PW in the membrane.

Note: Ethanol residues in Buffer PW can inhibit subsequent enzymatic reactions (restriction enzyme digestion, PCR, etc.) experiments.

Step 9



Transfer the Spin Column CR2 into a 1.5 ml centrifuge tube, and add 20-50 μl Buffer TB to the middle of the adsorption membrane. Placed at room temperature for 2 min and centrifuge at 12,000 rpm (~13,400×g) for 2 min to collect the solution into the centrifuge tube.