(DP302) TIANamp Bacteria DNA Kit

——Bacteria

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Experiment Preparation

1. Bacteria culture solution 1-5 ml (This experiment took Gram-negative bacterium as an example, and the pre-treatment of gram-positive bacterium was described in the instruction.)
2. Pipette and matched sterile tips (10 μl, 200 μl, 1 ml); 1.5 ml centrifuge tubes
3. 96-100% ethanol
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.

Step 1

Take 1-5 ml bacteria culture solution, centrifuge at 10,000 rpm (~11,500×g) for 1 min and remove the supernatant. Add 200 μl Buffer GA to the bacteria pellet and vortex until the bacteria is completely suspended.

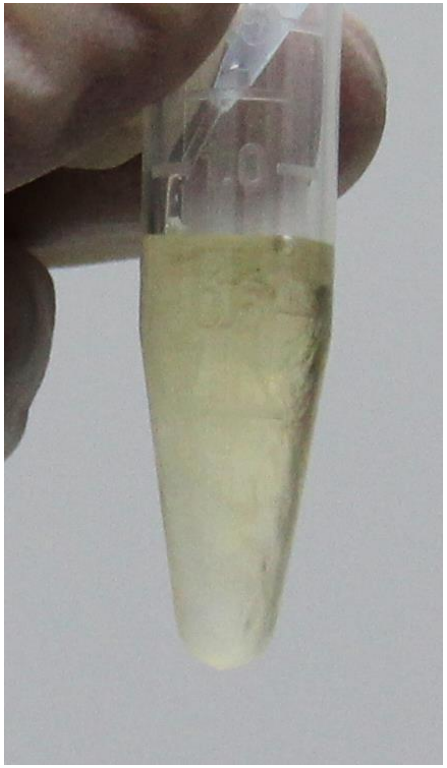
Step 2

Add 220 μl Buffer GB and 20 μl Proteinase K, and mix thoroughly upside down.

Step 3

Place the tube at 70℃ for 2 min, and solution should be clear. Briefly centrifuge to remove water drops of the cap and inner wall.

Step 4



Add 220 μl 96-100% ethanol, fully mix by vortex for 15 sec, and there may be flocculent precipitate at this time. Briefly centrifuge to remove water drops of the cap and inner wall.

Step 5

Transfer the solution and flocculent precipitate from the previous step to Spin Column CB3.

Centrifuge at 12,000 rpm (~13,400×g) for 30 sec, discard the waste liquid in the collection tube and place the Spin Column CB3 into the collection tube.

Step 6

Centrifuge at 12,000 rpm (~13,400×g) for 30 sec, discard the waste liquid in the collection tube and place the Spin Column CB3 into the collection tube.

Add 500 μl Buffer GDB to Spin Column CB3

Step 7

Add 600 μl Buffer PW to Spin Column CB3 (ensure 96-100% ethanol has been added before use).

Centrifuge at 12,000 rpm (~13,400×g) for 30 sec, discard waste liquid in the collection tube and place the Spin Column CB3 into the collection tube.

Step 8 Repeat step 7.

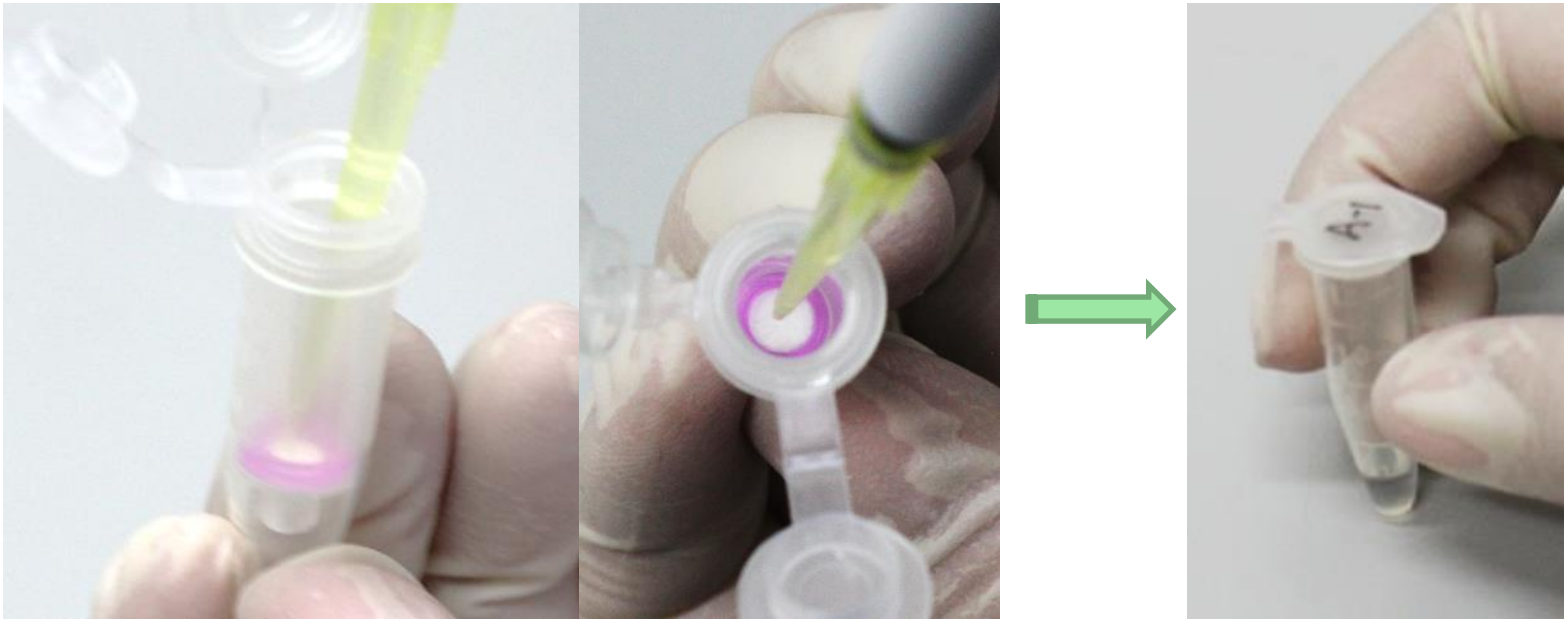
Step 9

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

Place the Spin Column CB3 at room temperature for 2 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 10



Transfer the Spin Column CB3 into a 1.5 ml centrifuge tube, and add 50-200 μl Buffer TE to the middle of the adsorption membrane. Place 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.