(DP331) TIANamp FFPE DNA Kit

——FFPE

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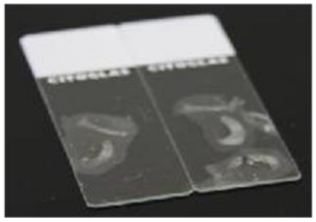
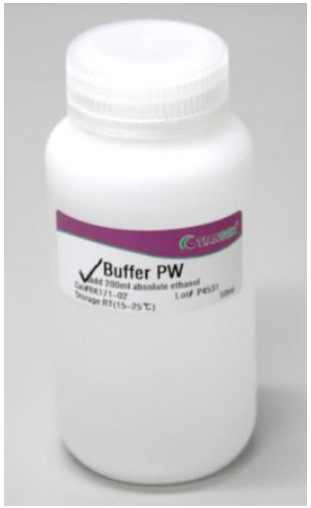
Experiment Preparation

1. Paraffin section or block
2. 96-100% ethanol; Xylene
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.

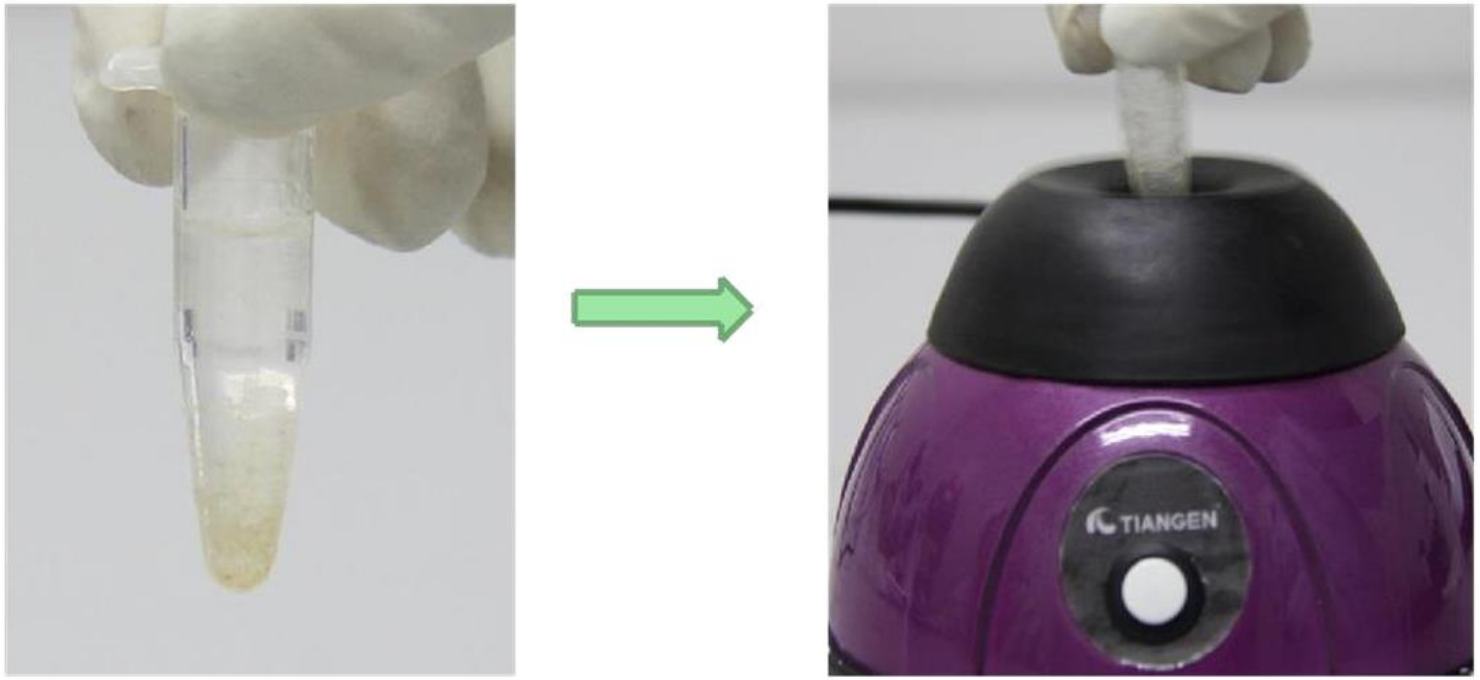


Step 1

Take 5-8 pieces of paraffin sections (5-10 μm,1×1 cm^2).

Note: If the surface of the sample is exposed to air, the first 2~3 scraped pieces shall be discarded.

Step 2



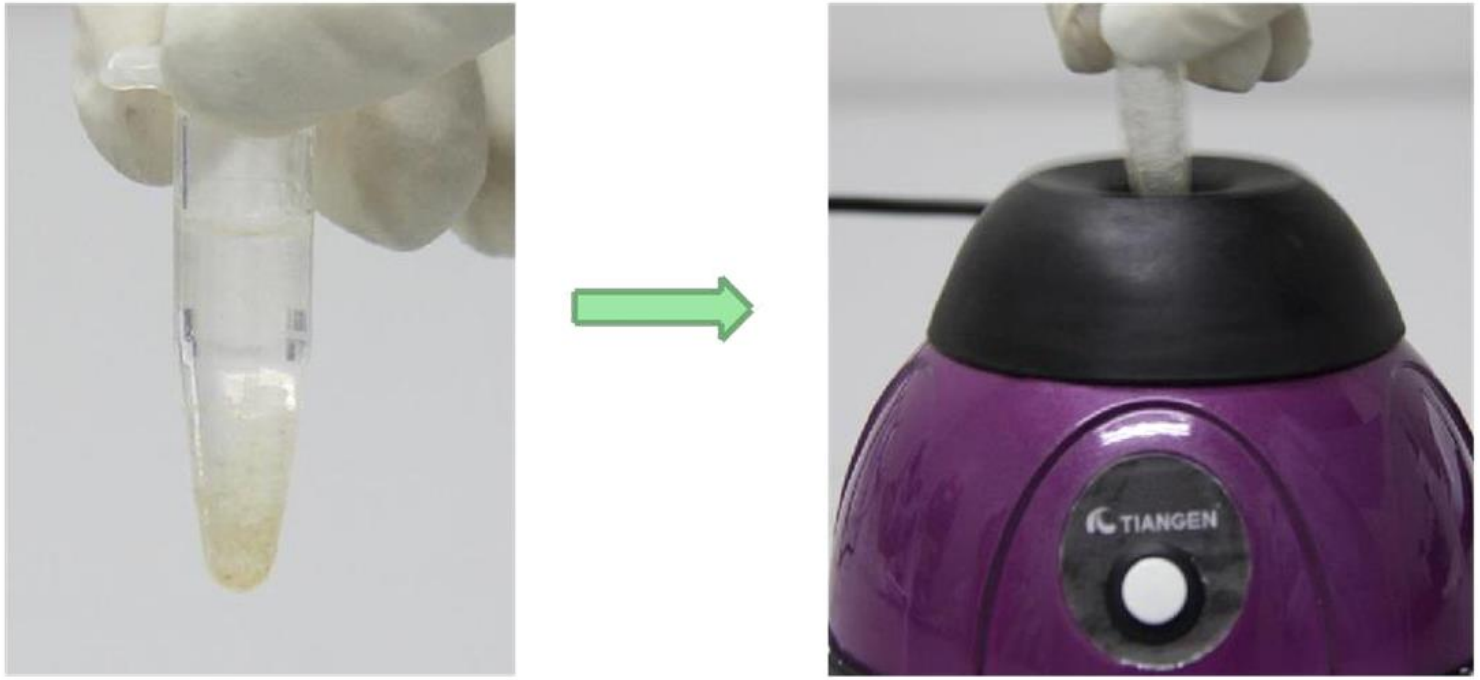
Place the sample in a 1.5 ml sterile centrifuge tube, add 1ml xylene, and vortex violently for 10 sec.

Step 3

Centrifuge at 12,000 rpm (~13,400 g) at room temperature for 2 min, discard supernatant.

Note: Do not pour out the precipitate (this step can be operated by a pipette).

Step 4



Add 1ml 96-100% ethanol, and mix thoroughly by vortex for 10 sec.

Step 5

Centrifuge at 12,000 rpm (~13,400 g) at room temperature for 2 min, discard supernatant.

Note: Do not pour out the precipitate (this step can be operated by a pipette).

Step 6

Place at room temperature for 5-10 min to fully volatilize ethanol.

Step 7

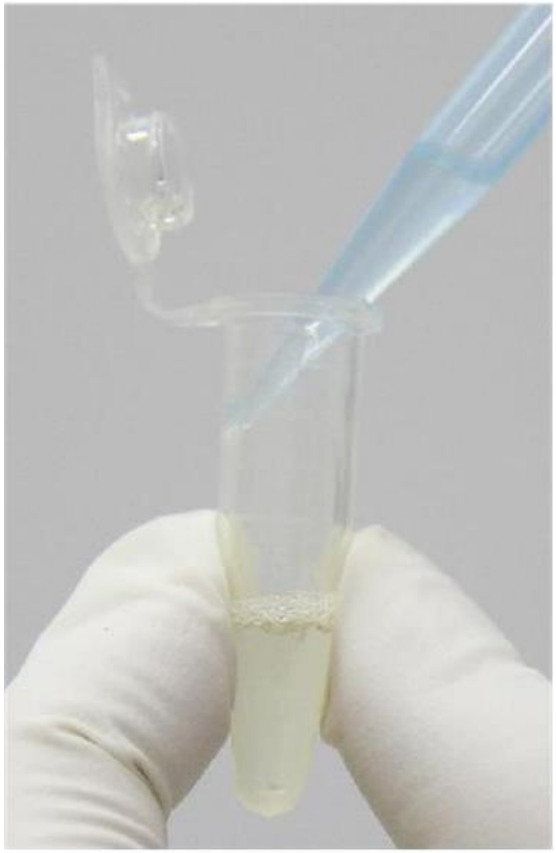
Add 200 µl Buffer GA and 20 µl Proteinase K.

Mix well, incubate for 1 h at 56 ℃ until the sample is completely lysed.

Step 8

Incubate at 90 ℃ for 1 h.

Step 9



Add 220 µl Buffer GB, mix well by vortex. Add 250 µl 96-100% ethanol, vortex to mix thoroughly.

Brief centrifuge to collect the drop on tube cap and wall to the bottom of the tube.

Step 10

Transfer the solution and flocculent precipitate from the previous step to Spin Column CR2.Centrifuge at 8,000 rpm（~6,000×g）at room temperature for 2 min, discard the waste liquid in the collection tube and place the Spin Column CR2 into the collection tube.

Step 11

Add 500 μl Buffer GD to Spin Column CR2.

Centrifuge at 8,000 rpm（~6,000×g）at room temperature for 1 min, discard the waste liquid and place the Spin Column CR2 into the collection tube.

Step 12

Add 600 μl Buffer PW to Spin Column CR2.

Centrifuge at 8,000 rpm（~6,000×g）at room temperature for 1 min, discard the waste liquid and place the Spin Column CR2 into the collection tube.

Step 13 Repeat step 12.

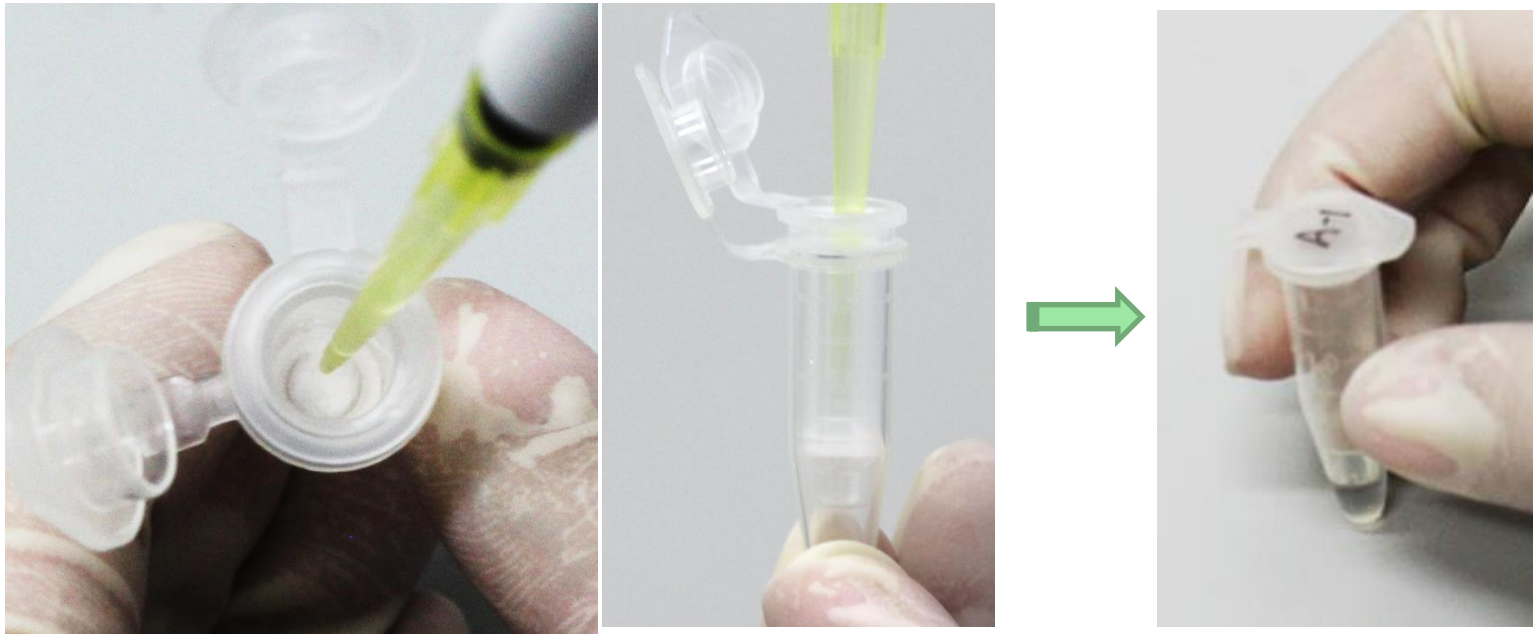
Step 14

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 15



Transfer the Spin Column CR2 into a 1.5 ml centrifuge tube, and add 65℃ preheated 30-100 μl Buffer TE or ddH_2O in to the middle of the adsorption membrane. Placed at room temperature for 2-5 min and centrifuge at 12,000 rpm (~13,400×g) for 2 min to collect the solution into the centrifuge tube. The purified DNA can be stored at -20℃.