TIANamp Blood Spots DNA Kit

Extraction of genomic DNA from dried blood spots samples

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TIANamp Blood Spots DNA Kit

(Spin Column)

Cat.no. GDP334

Kit Contents

Contents	GDP334-02 (50 preps)	GDP334-03 (200 preps)
Buffer GA	15 ml	50 ml
Buffer GB	15 ml	50 ml
Buffer GD	13 ml	52 ml
Buffer PW	15 ml	50 ml
Buffer TB	15 ml	30 ml
Proteinase K	1 ml	4 × 1 ml
RNase-Free Spin Columns CR2	50	200
Collection Tubes 2 ml	50	200
Centrifuge Tubes 1.5 ml	50	200
Handbook	1	1

Storage

This kit can be store at room temperature (15-30°C) for 15 months. If any precipitate forms in the buffers, it should be dissolved by warming the buffers at 37°C before use.

Introduction

The kit adopts a centrifugal adsorption column capable of specifically binding DNA and a unique buffer system to extract genomic DNA in dried blood spots. The silica matrix material used in the centrifugal adsorption column is a unique new material of TIANGEN, which can adsorb DNA efficiently and specifically, and can remove impurity proteins and other organic compounds in cells to the greatest extent. The extracted genomic DNA has large fragments, high purity and stable and reliable quality.

The DNA purified by the kit is suitable for various conventional operations, including enzyme digestion, PCR, fluorescence quantitative PCR, library construction, Southern blot, etc.

Sample Collection, Storage and Transportation

1. Sampling: Collect the sample according to the method of collecting dried blood spots with filter paper.
2. Sample storage: The sample collected above can be immediately used for processing, or stored under the conditions of sealing, drying (humidity lower than 30%) and 2 ~ 8°C for 5 years. When transporting samples, filter paper dried blood spots should be sealed and transported with foam box filled with ice.

Important Notes <u>Please read these notes before using this kit.</u>

1. If there is precipitation in Buffer GA or GB, it can be redissolved in a 37°C water bath and used after shaking to mix thoroughly.
2. After using the solution, tighten the bottle cap to avoid evaporation.
3. Perform all centrifugation steps at room temperature.
4. If the solution comes into contact with skin and mucous membrane, please immediately flush it with tap water. It will not cause harm to the operator.

Protocol

Before use, please add ethanol (96-100%) to Buffer GD and Buffer PW according to the instruction on the bottle label.

1. Take three 3 × 3 mm dried blood spot samples into a 1.5 ml centrifuge tube (self-provided).
2. Add 200 µl of Buffer GA to the tube.
3. Add 20 µl Proteinase K, vortex for 10 sec to mix well, then put it into a thermostatic shaker preheated to 56°C and shake at 900 rpm for 1 h.
4. Centrifuge briefly, add 200 µl of Buffer GB, vortex for 10 sec to mix well. Place the centrifuge tube into a thermostatic shaker preheated to 70°C and shake at 900 rpm for 10 min. After the incubation, centrifuge briefly to remove the droplets on the inner wall of the tube cover.

   Note: White precipitate may be generated when adding Buffer GB, which will disappear when it is placed at 70°C without affecting subsequent experiments. If the solution is not clear, it indicates that the cell lysis is not complete, which may lead to low DNA yield and purity.
5. Add 100 µl of ethanol (96-100%) to the tube. If the room temperature exceeds 25°C, precool the ethanol on ice. Gently invert to mix the sample, place it at room temperature for 5 min, and centrifuge briefly to remove the droplets on the inner wall of the tube cover.
6. Add the solution obtained in the previous step to a RNase-Free Spin Columns CR2 (place CR2 in a collection tube), centrifuge at 12,000 rpm (~ 13,400 × g) for 30 sec, discard the waste liquid from the collection tube, and put back the RNase-Free Spin Columns CR2 into the collection tube.
7. Add 500 µl Buffer GD to RNase-Free Spin Columns CR2 (**ensure that ethanol has been added before use**), centrifuge at 12,000 rpm (~ 13,400 × g) for 30 sec, discard waste liquid from the collection tube, and return RNase-Free Spin Columns CR2 to the collection tube.
8. Add 700 µl of Buffer PW to the RNase-Free Spin Columns CR2 (**ensure that ethanol has been added before use**), centrifuge at 12,000 rpm (~ 13,400 × g) for 30 sec, discard the waste liquid from the collection tube, and return the RNase-Free Spin Columns CR2 to the collection tube.
9. Add 500 µl of Buffer PW to RNase-Free Spin Columns CR2 and centrifuge at 12,000 rpm (~ 13,400 × g) for 30 sec. Discard the waste liquid from the collection tube.

10. Put the RNase-Free Spin Columns CR2 back into the waste liquid collection tube, centrifuge at 12,000 rpm (~13,400 × g) for 2 min, and discard the waste liquid. Place the RNase-Free Spin Columns CR2 at room temperature for 2-5 min to completely dry the residual buffer in the adsorption membrane.

    Note: The purpose of this step is to remove the residual buffer in the adsorption column. The residual ethanol in the buffer will affect the subsequent enzyme reaction (enzyme digestion, PCR, etc.).

11. Transfer the RNase-Free Spin Columns CR2 into a clean centrifuge tube, pipette 20-50 µl of Buffer TB in the middle of the adsorption membrane, place at room temperature for 2-5 min, centrifuge at 12,000 rpm (~ 13,400 × g) for 2 min, and collect the solution into the centrifuge tube.

    Note: The volume of the buffer should not be less than 20 µl. Too small a volume will affect the recovery efficiency. In order to increase the yield of genomic DNA, the DNA solution obtained by centrifugation can be added to adsorption column CR2 again, and placed at room temperature for 2 min, and centrifuged at 12,000 rpm(~13,400 × g) for 2 min. The pH value of the elute buffer has a great influence on the elution efficiency. If water is used as eluent, the pH value should be ensured to be within the range of 7.0-8.5 (the pH value of water can be adjusted to this range by NaOH), and the pH value lower than 7.0 will reduce the elution efficiency. DNA products should be kept at -20°C to prevent DNA degradation.