TIANamp Genomic DNA Kit

For isolation of genomic DNA from blood, cells and animal tissues

TIANGEN®

www.tiangen.com/en

TIANamp Genomic DNA Kit

(Spin Column)

Cat. no. DP304

Kit Contents

Contents	DP304-02 50 preps	DP304-03 200 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 TE	15 ml	60 ml
Proteinase K	1 ml	4×1 ml
Spin Columns CB3	50	200
Collection Tubes 2 ml	50	200
Handbook	1	1

Compatible Reagents

Red Cell Lysis Buffer (Cat. no. RT122);
RNaseA (100 mg/ml) (Cat. No. RT405-11)

Storage

TIANamp Genomic DNA Kit can be stored dry at room temperature (15-25°C) for up to 12 months without showing any reduction in performance and quality. For longer storage, the kit can be stored at 2-8°C. If a precipitate has formed in Buffer under 2-8°C, please place the buffer at room temperature or warm at 37°C for 10 min to dissolve the precipitate.

Introduction

TIANamp Genomic DNA Kit is based on silica membrane technology and provides special buffer system for many kinds of sample's gDNA extraction. The spin column is made of new type silica membrane can bind DNA optimally on given salt and pH conditions. Simple centrifugation processing completely removes contaminants and enzyme inhibitors such as proteins and divalent cations. Purified DNA is eluted in low-salt buffer or water, ready for use in downstream applications.

DNA purified by TIANamp Genomic DNA Kit is highly suited for restriction analysis, PCR analysis, Southern blotting, and cDNA library.

Yield

Source	DNA Yield
Whole blood from mammalian (100-400 µl)	3-10 µg
Whole blood from bird or amphibian (5-20 µl)	5-40 µg
Cultured cells (10^6-10^7 cells)	5-30 µg
Tissue (30 mg)	10-30 µg

Important Notes

1. Please add ethanol (96-100%) to buffer GD and Buffer PW as indicated on the bottle for the first use.
2. Repeated freezing and thawing of stored samples should be avoided, since this leads to reduced DNA size.
3. If a precipitate has formed in Buffer GA or Buffer GB, warm buffer to 37°C until the precipitate has fully dissolved.
4. All centrifugation steps should be carried out in a conventional table-top microcentrifuge at room temperature (15-25°C).

Protocol

Ensure that Buffer GD and Buffer PW have been prepared with appropriate volume of ethanol (96-100%) as indicated on the bottle and shake thoroughly.

1. Samples preparation

a. For blood, please use 200 µl fresh, frozen or anticoagulant-adding blood. If less than 200 µl, please make up with buffer

GA to 200 µl.

Note: If the blood volume is 300 µl-1 ml, please refer to the following step: add 3 times volume of Red Cell Lysis Buffer (Cat. No. RT122) to the sample (e.g., add 900 µl Red Cell Lysis Buffer to 300 µl blood), then close the cap and invert the tube. Stay the tube in room temperature (15-25°C) for 5 min, and centrifuge at 12,000 rpm (~13,400 × g) for 1 min, then discard the flow-through and pipet 200 µl buffer GA and mix by pulse-vortex.

b. If the sample is blood from poultry, birds, amphibians, of which red blood cells have nucleolus, the amount should be reduced to 5-20 µl and adjust the volume to 200 µl with buffer GA.

c. The adherent cells should be treated to cell suspension first, then centrifuge the cells for 1 min at 10,000 rpm (~11,200 × g), then discard the flow-through and re-suspend cell pellet in 200 µl buffer GA.

d. Animal tissue (spleen<10 mg) should be treated to cells suspension first, then centrifuge at 10,000 rpm (~11,200 × g) for 1 min , then discard the flow-through and re-suspend cell pellet in 200 µl buffer GA.

Note: If RNA-free genomic DNA is required, add 4 µl RNase A (100 mg/ml, should be prepared by user, Cat. No. RT405-12). Mix by vortex for 15 s, and incubate for 5 min at room temperature (15-25°C).

2. Add 20 µl Proteinase K, mix thoroughly by vortex.
   If the sample is tissue: incubate at 56°C until the tissue is completely lysed.

Note: Lysis time varies depending on the type of tissue processed. Lysis usually takes 1-3 h (rat tail needs to be lyzed overnight). Samples should be inverted 2-3 times every one hour. Or use Shaking Water Bath.

3. Add 200 µl Buffer GB to the sample, mix thoroughly by vortex, and incubate at 70°C for 10 min to yield a homogeneous solution. Briefly centrifuge the 1.5 ml microcentrifuge tube to remove drops from the inside of the lid.

Note: White precipitates may form when Buffer GB is added. They will not interfere with the procedure and will dissolve during the heat incubation at 70°C. If precipitates do not dissolve during heat

incubation, it indicates that the cell is not completely lysed and may result in low yield of DNA and impurity in DNA.

4. Add 200 µl ethanol (96-100%) to the sample, and mix thoroughly by vortex for 15 s. A white precipitate may form on addition of ethanol. Briefly centrifuge the 1.5 ml microcentrifuge tube to remove drops from the inside of the lid.
5. Pipet the mixture from step 4 into the Spin Column CB3 (**in a 2 ml collection tube**) and centrifuge at 12,000 rpm (~13,400 × g) for 30 s. Discard flow-through and place the spin column into the collection tube.
6. Add 500 µl Buffer GD (**Ensure ethanol (96-100%) has been added**) to Spin Column CB3, and centrifuge at 12,000 rpm (~13,400 × g) for 30 s, then discard the flow-through and place the spin column into the collection tube.
7. Add 600 µl Buffer PW (**Ensure ethanol (96-100%) has been added**) to Spin Column CB3, and centrifuge at 12,000 rpm (~13,400 × g) for 30 s. Discard the flow-through and place the spin column into the collection tube.
8. Repeat Step 7.
9. Centrifuge at 12,000 rpm (~13,400 × g) for 2 min to dry the membrane completely.

Note: The residual ethanol of buffer PW may have some affection in downstream application.

10. Place the Spin Column CB3 in a new clean 1.5 ml microcentrifuge tube, and pipet 50-200 µl Buffer TE directly to the center of the membrane. Incubate at room temperature (15-25°C) for 2-5 min, and then centrifuge for 2 min at 12,000 rpm (~13,400 × g).

Note: If the volume of eluted buffer is less than 50 µl, it may affect recovery efficiency. The pH value of eluted buffer will have a great effect on eluting, we suggest using buffer TE or distilled water (pH 7.0-8.5) to elute gDNA. For long-term storage of DNA, eluting in Buffer TE and storing at -20°C is recommended, since DNA stored in water is subject to acid hydrolysis.

Ordering Information

Genomic DNA Purification

Product	Size	Cat. no.
TIANamp Blood DNA Kit	50 preps	DP318-02
	200 preps	DP318-03

RNA Purification

Product	Size	Cat. no.
RNAprep Pure Kit for Plant	50 preps	DP432

PCR MasterMix

Product	Size	Cat. no.
2x *Taq* PCR MasterMix (with loading dye)	1 ml	KT201-01
	5 x 1 ml	KT201-02
2x *Taq* Plus PCR MasterMix (with loading dye)	0.5 ml	KT205-01
	5 x 1 ml	KT205-02

DNA Marker

Product	Size	Cat. no.
DNA Marker I	50 lanes	MD101-01
	200 lanes	MD101-02