AllPure Blood Genomic DNA Kit

Cat. No. ABTGNA024

Storage:RNase A and Proteinase K solutions at -20°C for one year; others at room temperature (15-25°C) for one year

Description

AllPure Blood Genomic DNA Kit provides a simple and convenient way to isolate high quality genomic DNA from 5-250 µl of fresh or frozen blood. Whole blood is incubated with binding/lysis buffer to release DNA. DNA is bound to silica-based column.

The isolated DNA is suitable for PCR, restriction enzyme digestion and Southern blot.

- Simple and fast, red cell lysis buffer is no longer needed.
- Complete removal of contaminants and inhibitors.
- DNA yield up to 40 µg.
- Column based purification, no organic extraction or ethanol precipitation.
- Suitable for EDTA, sodium citrate and heparin-anticoagulated fresh or frozen blood in a volume of 5 to 250 µl.

Starting material

For blood with nonnucleated erythrocytes, we suggest using 5-250 µl; others, we suggest using 5-20 µl.

Kit Contents

Component	50 rxns	200 rxns
Binding Buffer 3 (BB3)	30 ml	110 ml
Clean Buffer 3 (CB3)	6 ml	24 ml
Wash Buffer 3 (WB3)	12 ml	2×22 ml
Elution Buffer (EB)	25 ml	80 ml
RNase A (20 mg/ml) -20°C	500 µl	2×1 ml
Proteinase K (20 mg/ml) -20°C	1 ml	4×1 ml
Genomic Spin Columns with Collection Tubes	50 each	2×100 each

Procedures

Before starting, add the indicated volume of 96%-100% ethanol into the concentrated CB3 and WB3.

Components	50 rxns	200 rxns
Clean Buffer 3 (CB3)	24 ml	96 ml
Wash Buffer 3 (WB3)	48 ml	2×88 ml

All centrifugation steps are carried out at room temperature.

1. Add the appropriate volume of blood, 20 µl of Proteinase K and 500 µl of BB3 into a microcentrifuge tube. Mix for 15 seconds by vortexing, and then incubate at room temperature for 10 minutes.
   Optional: If RNA-free genomic DNA is required, add 20 µl of RNase A before incubation.
2. Centrifuge briefly, add all the lysate to a spin column. centrifuge at 12,000×g for 1 minute, discard the flow through.
3. Add 500 µl of CB3 (check to make sure ethanol has been added), centrifuge the tube at 12,000×g for 30 seconds, discard flow through.
4. Add 500 µl of WB3 (check to make sure ethanol has been added), centrifuge the tube at 12,000×g for 30 seconds, discard flow through.

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5. Repeat step 4 once.
6. Place the spin column to a collection tube. Centrifuge the empty column at 12,000×g for 2 minutes to remove any residual WB3. Air-dry the spin column at room temperature for several minutes.
7. Place the spin column in a sterile 1.5 ml microcentrifuge tube. Add 50-200 µl of Elution Buffer (for higher yield, prewarm the buffer to 60°C) or distilled water (pH >7.0) to the center of column. Incubate at room temperature for 1 minute. Centrifuge at 12,000×g for 1 minute to elute the genomic DNA (to recover more DNA, add Elution Buffer or distilled water again to perform a second elution). For long-term storage, store the purified DNA at -20°C

Notes

• It is important not to overload the column, as this can lead to significantly lower yields than expected.
• Use fresh material and avoid repeated thawing and freezing.
• Use sterile tubes and pipette tips to avoid the contamination from DNase.