

PrimePrep™ Genomic DNA Extraction Kit (from Blood)

Introduction

PrimePrep™ Genomic DNA Extraction Kit (from Blood) is designed for the rapid preparation of genomic DNA from up to 100 µl of a blood sample. This kit is suitable to use with whole blood treated with either citrate or EDTA. It takes about 20 minutes to complete all processing of samples.

The purified genomic DNA is fully digestible with all restriction enzymes tested, and is completely compatible with PCR and Gene analysis.

Storage Conditions and Product Stability

All solutions should be tightly sealed and stored at room temperature. These reagents should remain stable for at least 1 year in their unopened containers.

The Proteinase K solution (20 mg/ml) should be stored at -20 °C.

Kit Components

Reagents \ Cat.No.	K-2000 (50 prep.)	K-2001 (200 prep.)
Spin column	50 ea	50 ea x 4
Collection tube	100 ea	100 ea x 4
Buffer GB	12 ml	12 ml x 4
Buffer GW1	20 ml	20 ml x 4
Buffer GW2	10 ml	10 ml x 4
Buffer GE	10 ml	10 ml x 4
Proteinase K Sol. (20 mg/ml)	1.2 ml	1.2 ml x 4

Before you begin

1. Add 15 ml ethanol to Buffer GW1 before use.
2. Add 40 ml ethanol to Buffer GW2 before use.
3. Check Buffer GB and GW1 before use for salt precipitation.

Note: Redissolve any precipitation by warming to 50 °C.

Experimental Protocol

1. Add 20 μ l of Proteinase K solution (20 mg/ml) to a 1.5 ml microcentrifuge tube.
2. Transfer 200 μ l of sample (whole blood, plasma, serum, buffy coat and body fluids) to the microcentrifuge tube.
If the sample volume is less than 200 μ l, add the appropriate volume of PBS.
3. **Optional RNase A treatment** If RNA-free genomic DNA is required, add the 20 μ l of RNase A Solution (10 mg/ml).
4. Add 200 μ l of Buffer GB to the sample. Mix well by pulse-vortexing for 15 sec.
5. Incubation at 56°C for 10 minutes.
Longer incubation times have no effect on yield or quality of the purified DNA.
6. Add of 200 μ l absolute ethanol and mix well by pulse-vortexing for 15 sec.
After this step, briefly spin down to get the drops clinging under the lid.
7. Carefully transfer the lysate into the upper reservoir of the spin column (fit in a 2 ml tube) without wetting the rim.
8. Centrifuge at 10,000 rpm for 1 min.
9. Transfer the spin column to a new 2 ml collection tube for filtration.
10. Add 500 μ l of Buffer GW1 to the spin column, and centrifuge at 10,000rpm for 1min.
11. After centrifugation, discard the flowthrough and transfer the spin column to a new 2 ml collection tube.
12. Add 500 μ l of Buffer GW2 to the spin column, and centrifuge at 10,000rpm for 1min.
13. After centrifugation, discard the flowthrough and reassemble the spin column with its collection tube.
14. Centrifuge once more at 12,000 rpm for 1~2 min to completely remove ethanol, and check that there is no droplet clinging to the bottom of collection tube.
Residual GW2 in the spin column may cause problems in later application.
15. Transfer the spin column to a new 1.5 ml tube (not provided) for elution. Add 200 μ l of Buffer GE onto spin column, and wait for at least 1 min at room temperature.
16. Elute DNA by centrifugation at 10,000 rpm for 1 min.