



## Genorise® Blood DNA Extraction Kit 500

This kit is to isolate total DNA including genomic and mitochondrial DNA from 0.3 ml of blood and is for 500 applications. If a smaller or larger sample volume is employed, the reagent quantity should be proportionally decreased or increased. This kit can significantly quality and quantity of DNA and is much cheaper than the similar products (\$0.32/application).

### Materials provided in the kit:

100 ml 10 x Red Blood Cell Lysis Solution  
125 ml 5 x Lysis Solution  
120 ml Protein Precipitation Solution  
200 ml DNA Hydration Solution

### Materials required but not provided in the kit:

Isopropanol (2-propanol)/Ethanol

## Protocol

### Cell Lysis

1. Dilute 10 x Red Blood Cell Lysis Solution to 1 x by DNA water.
2. Take 0.3 ml whole blood to a 1.5 ml microcentrifuge tube, add 0.9 ml 1 x Red Blood Cell Lysis Solution, vortex briefly, and incubate for 2 minutes to lyse the red blood cells.
3. Centrifuge 1 min at 13,000 x g to pellet the white blood cells and discard the supernatant by a pipette (repeat red blood cell lysis if cell pellet is red), leaving 20 µl liquid residue, and resuspend the white cells by a pipette.
4. Add 0.24 ml DNA water and 0.06 ml of diluted 5 x Cell Lysis Solution, resuspend cells by a pipette.
5. Incubate at 55°C for 1 hr to overnight until cell lysate becomes completely clear (disrupted).

### Protein Precipitation

1. Cool sample to room temperature by placing on ice for 1 min.
2. Add 0.1 ml of Protein Precipitation Solution to the lysate and vortex samples for 20 sec.
3. Place sample into an ice bath for 5 min.
4. Centrifuge at 15,000 x g for 5 min.

### DNA Precipitation

1. Pour the supernatant containing DNA into a new 1.5 ml microcentrifuge tube.
2. Centrifuge at 15,000 x g for 5 min.
3. Pour the supernatant to a new microcentrifuge tube; repeat step 1 and 2 until no pellet is seen.
4. Pour the supernatant containing DNA into a new 1.5 ml microcentrifuge tube containing 0.3 ml 100% isopropanol.
5. Mix the samples by inverting gently 50 times and incubate at room temperature for 5 min.
6. Centrifuge at 13,000 x g for 5 min.
7. Pour off the supernatant and drain the tube briefly on clean absorbent paper. Add 1 ml of 70% ethanol and invert the tube several times to wash the DNA pellet.
8. Centrifuge at 13,000 x g for 5 min, carefully pour off the ethanol and do not lose the DNA pellet.
9. Invert tube and drain the tube on clean absorbent paper, remove the remaining liquid by a pipette, and finally allow air-drying for 5 min.



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### **DNA Hydration**

1. Add 50  $\mu$ l DNA Hydration Solution.
2. Resuspend the DNA pellet by a pipette for 5 times and rehydrate the DNA by incubation for 10 min at room temperature.
3. Vortex briefly and pulse spin before use, and store at  $-20^{\circ}\text{C}$ .

### **Note**

1. Following DNA precipitation, if you see a big pellet with a color other than white, we recommend you repeat this protocol and 1 hr cell lysis.
2. Air drying of DNA pellet should not exceed 5 min.