Genorise Plasmid DNA Extraction Kit 200

This kit is to isolate plasmid DNA from 1 ml cultured E. coli containing plasmid DNA and is for 200 applications. This kit can significantly improve quantity and quality of DNA and is much more cost-effective than the similar products.

Materials provided in the kit:
100 ml Lysis Solution
40 ml Protein Precipitation Solution
50 ml DNA Hydration Solution
0.7 ml 10 x DNA Sample Loading Buffer

Materials required but not provided in the kit:
Isopropanol (2-propanol)/Ethanol

Protocol

Cell Lysis

1. Add 1 ml E. coli cell suspension containing plasmid DNA from an overnight culture to a 1.5 ml microcentrifuge tube.
2. Centrifuge at 13,000 x g for 10 seconds to pellet the cells. Carefully remove as much supernatant as possible with a pipet.
3. Add 0.5 ml of Lysis Solution to the E. coli pellet, vortex for 20 sec.
4. Incubate at 55°C for 1 hr or overnight until the cell lysate becomes completely clear (disrupted).

Protein precipitation

1. Cool sample to room temperature by placing on ice for 1 min.
2. Add 0.2 ml of Protein Precipitation Solution to the lysate.
3. Vortex samples at high speed for 20 sec and 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 1.7 ml microcentrifute 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 0.5 ml 100% isopropanol (2-propanol).
5. Mix the samples by inverting gently 50 times and incubate at room temperature for 10 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 and drain the tube on clean absorbent paper, completely remove the remaining liquid by a pipette, and finally allow airing dry for 5 min.

DNA Hydration

1. Add 50 µl DNA Hydration Solution.
2. Resuspend the DNA 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°C.

Note

1. Following DNA precipitation, if you see a big pellet with a color other than white, we recommend you repeat this protocol with a start volume of 0.5 ml of DNA isolate and 1 hr cell lysis.
2. Do not air dry DNA pellet more than 5 min.