

# Genorise® Plasmid DNA Extraction Mini Kit 100

This kit uses mini-DNA binding column to isolate plasmid DNA from 1-5 ml of plasmid DNA-containing E coli and is for 100 applications. This kit can significantly increase quality and quantity of DNA and is more cost-effective than the similar products.

## Materials provided in the kit:

- 1. 100 mini–DNA Binding columns
- 2. 100 Collection tubes
- 3. 60 ml Buffer GB
- 4. 30 ml Buffer GS
- 5. 30 ml Buffer GL
- 6. 40 ml Buffer GP
- 7. 28 ml Buffer GW1
- 8. 18 ml Buffer GW2
- 9. 12 ml Buffer GE

## Materials required but not provided in the kit:

- 1. 100% Ethanol
- 2. 1.5 ml microcentrifuge tube

### Reagent preparation

1. Buffer GW1: add 32 ml 100% ethanol

2. Buffer GW2: add 42 ml 100% ethanol

### **Protocol**

- 1. Add 500 μl Buffer GB to DNA binding column with collection tube, centrifuge 13,000 x g for 1 min and discard the flowthrough.
- 2. Centrifuge 1-5ml bacterial culture at 13,000 x g for 1 min and discard supernatant. Pipet 250 μl Buffer GS into pellet and suspend bacterial cells and transfer to a 1.5 ml microcentrifuge tube.
- 3. Add 250 µl of Buffer GL and mix thoroughly by inverting the tube until solution becomes clear. Cell lysis should be done in 5 min.
- 4. Add 350 μl Buffer GP and mix thoroughly by inverting the tube for 4-6 times. Centrifuge at 16, 000 x g for 5 min.
- 5. Pipet the supernatant onto the column with collection tube and centrifuge at full speed (20,000 x g) for 1 min. If samples remain in the column, repeat centrifugation to completely remove the remaining samples. Discard the flow-through andbut keep collection tube.
- 6. Add 500 μl Buffer GW1 to the column with collection tube and add 500 μl Buffer GW1. Centrifuge at 6000 x g for 1 min. Discard the flow-through and keep the collection tube.

- 7. Add 500 µl Buffer GW2. Centrifuge at full speed (20, 000 x g) for 3 min. Discard the flow-through and keep the collection tube.
- 8. Centrifuge at full speed for 1 min. Discard the flow-through and collection tube.
- 9. Place the mini column in a new 1.5 ml microcentrifuge tube and add 50-100  $\mu$ l Buffer GE and incubate for 2 min. Centrifuge at 6000 x g for 1 min to elute DNA.