The mass plasmid DNA extraction

Materials:

The mass plasmid DNA extraction kit (Dingguochangsheng,catalog no. NEP012-2)
Bacteria solution

Procedure:

1、Taking 150ml overnight culture of bacteria, centrifuge at 8000 RPM for 10 minutes, and abandon the clear liquid.

2、Add 8 ml of solution A to suspend bacterial precipitation and suspend evenly until there are no bacterial clumps.

3、Add 10ml solution B and stir gently and thoroughly for 3 minutes until the solution is clear and thick .

4、Add 10ml solution C, stir gently and thoroughly until a white flocculent precipitate forms, and let stand at room temperature for 5 minutes.

5、10000 RPM centrifuge for 10 min.

6、Transfer the clear liquid to beaker or centrifugal tube, add 0.3 times volume of isopropyl alcohol, mix well.

7、Transfer the solution to a centrifugal column and let stand for 2 minutes.

8、12000 RPM centrifuge for 10 min, and abandon the clear liquid (repeat this process three times).

9、Add 8ml solution D(add anhydrous ethanol before use), stand for 2 minutes, centrifuge at 10000 RPM for 1 minute, and discard the waste liquid.

10、Repeat step 9 once.

11、Add 3 ml anhydrous ethanol, 10000 RPM centrifugal 1 min.

12、Centrifuge again at 10000 RPM for 2 minutes to dry the remaining liquid.

13、Put centrifugal column in a clean centrifuge tube, open cover and let stand for 5-10 minutes to clean the residual ethanol. Fill the centrifuge column with 1-2ml EB or deionized water and stand at room temperature for 2 minutes.

14、12000 xg centrifugal 2 minutes, wash plasmid, save plasmid in - 20 ℃.