Protocol 1-7: Chemical Transformation of Recombinant DNA

General Protocol

1)Pipette competent cells suspension into the tubes, 100 μl each tube (If the competent cells are taken from -70℃, perform following steps immediately after thawing).

2)Add 10 μl of recombinant plasmid into the tube which is the tube with competent cells.

3)Mix the solutions gently, keep on ice for 20-30 minutes.

4)Heat shock by transferring the tubes to a water bath of 42℃ for 1 to 2 minutes.

5)Immediately return the tube to the ice bath. Keep on ice for 2 minutes.

6)Add 0.9 ml of LB (with no antibiotics added) into each tube. Incubate the tubes for 45 minutes to 1 hour at 37℃ to allow the cells to express their antibiotic gene product.

7)Spread about 200 μl of the resulting solutions (do dilution if necessary) on LB plates (with corresponding antibiotic added). After complete absorption of liquid LB, upside down the plates and incubate the plates at 37℃ overnight.

Tips

1)Never spread the transformation solution until you have assured that the glass stick is cooled down