PLATING & TITERING λ PHAGE

Preparation of plating cells.

1. Streak a colony of host cells on LB and incubate at 37°C overnight.

2. Inoculate 50 ml LB + 10 mM $MgSO_4$ + 2% maltose, incubate 4-6 h at 37°C or overnight at 30°C. Do not let OD_{600} exceed 1.

3. Pellet bacteria at 2000 rpm for 10'. Resuspend cells in half the original volume of sterile 10 mM $MgSO_4$.

4. Dilute cells to an OD of 0.5 with 10 mM $MgSO_4$. Store at 4°C and use within 48 h.

This is the correct way to do it, however, you get just as good results using an overnight culture that has been grown as in 2 and then stored at 4°C.

Plating.

1. Make dilutions of phage stock in SM.

2. Mix 200 µl plating cells with up to 100 µl phage solution. Incubate without shaking at 37°C for 15'.

3. Add 5 ml top agarose (LB + 0.7% agarose), mix well and pour over an LB plate. Incubate 37°C overnight.

4. Calculate titer on the basis of pfu (plaque forming units) per µl phage stock.

SM (100 ml)
2 ml 5 M NaCl
2.5 ml 2 M Tris pH 7.5
1 ml 1 M $MgCl_2$
500 µl 2% gelatin