PCR analysis of lambda phage

Judelson 9-97

1. Using a sterile pasteur pipette, carefully remove the desired plaque(s) from the plate. GENTLY expel the plaque into a 1.5 ml tube containing 200 µl of 1/2 strength SM. Add 20 µl chloroform and gently hit the tube with your fingers. Let sit 2 hr at room temp.

2. Remove 5 µl of the phage suspension and place in PCR tube containing 10 µl water. Heat 5 minutes at 98^0C.

3. Add 15 µl of 2X concentrated PCR reaction mix.

   Recipe for 2X mix (1 reaction)
   3 µl 1OX PCR buifer
   0.3 µl 100 x BSA stock (10 mg/mi)
   2.4 µl 1.25 mM dNTP solution
   0.24 µl Taq polymerase
   50 ngof each primer (0.05 µl of 1 mg/ml stock).

4. Do amplification with appropriate primer set. For the GT1O primers: use 50C anneaiing, 30-35 cycles.