Caesium Chloride Density Gradient Centrifugation

Introduction

How to purify the concentrated bacteriophage solution so that is ready for the immobilization onto the electrode.

Materials

- Caesium chloride (CsCl)
  - ρCsCl(aq)=1,6 g/ml
  - ρCsCl(aq)=1,5 g/mL
  - ρCsCl(aq)=1,4 g/mL
  - ρCsCl(aq)=1,3 g/mL
- 1x TM Buffer

Procedure

- Day 1

1. Fill the ultracentrifuge tube (25 mL PC Ultracentrifuge tube) with (starting with 1,6) :

   - 2 mL ρCsCl(aq)=1,6 g/ml
   - 3 mL ρCsCl(aq)=1,5 g/mL
   - 6 mL ρCsCl(aq)=1,4 g/mL
   - 6 mL ρCsCl(aq)=1,3 g/mL

2. Pipette 5 mL from the dissolved bacteriophage pellet on top of the tube

3. Centrifuge at 160,000 x g (46 700 rpm), 2.5 h, 4 °C

4. Pipette the phage band with a pipette

   - Phage band should be located between the 1.4 and 1.5 phase

5. Do an overnight dialysis, with a 2.000 x Vol 1x TM-Buffer in order to remove the caesium and chlorides from the purified phage solution