Mini Prep

The Mini Preparation is a method of extracting and purifying plasmid DNA. This protocol can be used for the precipitation of plasmid DNA from an 2 ml *E.coli* culture.

Materials

- Sol I
  - 50 mM Glucose(-monohydrate)
  - 10 mM EDTA
  - 5 mM Tris
  - H20
- Sol II
  - 0.5 M NaOH
  - 20% SDS
  - H20
- Sol III
  - 5M Potassium Acetate (60 ml)
  - Acetic Acid (11.5 ml)
  - H20 (to 100 ml)

- 100% Isopropanol
- Sodium Acetate
- 70% EtOH
- Tris-HCL

Procedure

- Centrifugation of 2 ml over night E.coli culture (2 min, 16.000 rpm, RT)
- Discard supernatant
- Resuspend pellet in 100 ul Sol I
- Add 150 ul Sol II and mix the contents by gently inverting the vessel several times
- Carry out all further steps on ice
- Add 200 ul of ice-cold Sol III and mix the contents by gently inverting the vessel several times
- Centrifuge the suspension for 2 min, 16.000 rpm, 4 °C
- Transfer the supernatant into new vessel, without transferring parts of the pellet
- Add 450 ul ice-cold Isopropanol and 45 ul sodium acetate
- Mix the contents by inverting the tube 20 times
- Centrifuge the suspension for 2 min, 16.000 rpm, 4 °C
- Discard the supernatant
- Wash the pellet twice 200 ul with 70% EtOH by inverting the tube several times and centrifuging for 1 min (16.000 rpm, RT)
- Dry the pellet at 37°C
- Dissolve DNA in 20-40 ul Tris-HCL