Rapid DNA Extraction from Cyanobacteria

Introduction

Get started by giving your protocol a name and editing this introduction.

Materials

- Phenol:Chloroform
- isopropanol
- 70% EtOH
- TE Buffer (10T/1E)
- 10% SDS
- lysozyme [50 mg/ml]
- 5M NaCl
- **Some serious protective gear!**

Procedure

1. Pellet the cells if liquid o/n culture. If you pick a colony from plate suspend it in TE buffer and pellet the cells.
2. Wash the pellet with TE buffer twice in order to remove any residual media.
3. Remove supernatant and add 500 µl TE buffer.
4. Add SDS so final conc. is 1% and ad 50 µl of lysozyme.
5. Keep at 70 degrees C for 15 min.
6. Add equal volume of phenol:chloroform and mix thouroughly.
7. Centrifuge at 10 000 rpm for 10 min.
8. Transfer supernatant to a new tube and ad equal volume of chloroform. Centrifuge at 10 000 rpm for 10 min in a temp. controlled centrifuge.
9. Repeat step 8.
10. Transfer supernatant to a new tube and ad 0.1 volume of 5M NaCl and ad 2 volumes of isopropanol. Keep at -20 degrees C for 2 hrs.
11. Centrifuge at 15 000 rpm for 30 min at 10 degree C.
12. Wash the pellet with 70% EtOH and centrifuge at 10 000 rpm for 10 min. Repeat.
13. Dry pellet and reuspend in 50 µl MilliQ.