

## Miniprep (alkaline lysis)

- solutions:
  - solution 1: 50 mM glucose  
10 mM EDTA  
25 mM Tris pH 8.0
  - solution 2: 0.2 N NaOH  
1% SDS
  - solution 3: 60 ml 5M KOAc  
11.5 ml 96% acetic acid  
28.5 ml H<sub>2</sub>O
  - TE: (from 10x TE)  
10x TE: 0.1 M Tris pH 8.0  
10 mM EDTA
- 1-5 ml E. coli over night culture
- spin down (eppendorf centrifuge)
- resuspend pellet in 100 µl solution 1 and add 2 µl RNaseA (10 mg/ml)  
(for 5 ml: 200 µl solution 1)
- add 200 µl solution 2 , mix by inversion (for 5 ml: 300 µl solution 2)  
(cells should lyse)
- leave at RT for 5 min, put on ice for 5 min
- add 150 µl solution 3 , mix carefully (for 5 ml: 300 µl solution 3)
- spin down for 30 min at 4°C (eppendorf centrifuge)
- transfer supernatant to new tube containing 1 vol. Isopropanol (450 µl)  
(for 5 ml: 800 µl isopropanol) , mix well
- spin down 20 min at RT (eppendorf centrifuge)
- remove supernatant, wash pellet 2x in 1 ml 70% EtOH (-20°C), spin 5 min
- dry pellet 5-10 min at RT
- dissolve in 25 µl 1x TE