

## Transformation – CaCl Method

(from Christopher)

- inoculate 50 µl from overnight culture (XL1 blue or DH5α) in 10 ml LB  
**(for 10 transformations)**
- let grow to  $OD_{600} = 0,4 - 0,6$  at 37°C with agitation (at least 200 rpm)
- prepare 0,2 M CaCl solution and **put it on ice!**  
(everything should be on ice all the time)
- centrifuge the cells at fullspeed for 1 min
- resuspend cells with 1 ml of 0,2 M CaCl solution
- use 100 µl cells for transformation
- add DNA (ca. 100 ng of plasmid)
- put it on ice for 30 min
- heat shock for 90 s at 42°C
- put it on ice again for 2 min
- add 400 µl of fresh LB
- let grow for 30 – 60 min at 37°C with agitation (at least 200 rpm)
- centrifuge the cells for 1 min and resuspend each tube with 100 µl LB
- plate on selective media