Heat Shock Transformation in E.coli

Name:

Date:

Project:

Heatshock Method!

- 1x 50µL Vial of chemical competent *E.coli* strain of interest per transformation
  - [ ] One Shot™ ccdB Survival™ 2 T1R
  - [ ] Top10
  - [ ] DB3.1
  - [ ] XL10-gold
  - [ ] SHuffle T7
  - [ ] SHuffle T7 express LysY
  - [ ] SHuffle express
  - [ ] SHuffle T7 express
  - [ ] BL21
  - [ ] BL21(DE3)
  - [ ] BL21(DE3) pLysS
  - [ ] Rosetta2(DE3)
  - [ ] ArcticExpress(DE3)
  - [ ] C43(DE3)

- [ ] 100ng of plasmid of interest per transformation

- [ ] 1x LB-Agar plate with desired selection marker per transformation

- [ ] S1 Benchtop Eppi Shaker
- [ ] incubator closet
- [ ] LB-Medium or SOC Medium

Day 1, date:

- select program 6 on S1 Benchtop Eppi Shaker
- get LB-Agar plate from cold room and leave on bench to get to roomtemp.
- get desired Plasmid from -20°C and thaw on ice
- get *E.coli* vial from -80°C and thaw on ice for 10min
- add 100ng of plasmid to 1x 50µL vial of *E.coli* competent cells
- mix by stirring gently with the pipette tip, do not vortex or mix by pipetting up and down
- chill on ice for 10min
- put vial into tabletop shaker and start program 6 (42°C, 45sec, no shaking)
- select program 7 on S1 Benchtop Eppi Shaker
- put vial back on ice until shaker has cooled to 37°C
- add _____µL LB or SOC Medium to vial (200 – 500µL)
- mix by pipetting
- put into shaker and start program 7 (37°C, 30min, 500 RPM)
- spin down cells in vial
- discard added LB Medium
- resuspend cells in remaining 50µL
- spread on prepared LB-Agar plate with desired selection marker with the help of a drigalski spatula or inoculating loop
- turn upside down and incubate at _____°C for _____h (e.g. 37°C overnight, or 22°C over the weekend)

Day 2, date:

- check LB-Agar plates for colonies