Transformation to 3300 LG using electroporation

1. The day before, prepare a starter (3 ml of LB with 3µl of the relevant antibiotics) of the relevant strain.
2. On the day of the transformation, transfer 0.5ml of starter into 50 ml of LB with antibiotics (in an erlenmeyer) and let them shake in 37°C at 250 rpm until they reach O.D. 0.6.

Prepare ice and make sure the autoclaved didistilled water and 10% glycerol are on it!

Work with cold centrifuge.

3. Once they reach that O.D., transfer the LB into 50 ml flasks and centrifuge for 10 minutes at 4000rpm. Discard the supernatant.
4. Break the pellet on ice and add 10ml of cold autoclaved didistilled water. Vortex to make sure the pellet is completely suspended.
5. Centrifuge for 10 minutes at 4000rpm, and then discard supernatant.
6. Break the pellet and add 10ml of cold 10% glycerol solution. Vortex to make sure the pellet is completely suspended.
7. Centrifuge for 10 minutes at 4000rpm, and then discard supernatant.
8. Repeat steps 6-7 twice more (3 rounds of 10% glycerol).
9. After the last round, suspend the pellet in 1 ml of 10% glycerol. Keep the bacteria on ice.
10. Create a **1**ng/µl solution of the DNA you want to transform into the cells.
11. Add **1µl** of this solution to **100µl** of cells. Transfer everything into an electroporation cuvette.
12. Insert the cuvette into the electroporator, while making sure the bulge faces forward.
13. Electroporate in 2500v, 5ms.
14. Immediately add 1 ml of SOB while transferring the bacteria from the cuvette to a 14ml tube.
15. Put the tube in the shaker at 250 rpm, 37°C for 1 hour.
16. Plate the bacteria on two plates (per reaction tube) containing appropriate antibiotics (choose two different volumes to plate, according to your sample). Leave the plates overnight in the 37°C incubator.
17. Return within approximately 16 hours to check the plates. Wrap the ones you'd like to save with parafilm and put them in the refrigerator.