Plasmid Prep from Cultures (using QIAprep Spin Miniprep Kit)

1. Pick and individual colony from a plate and inoculate 2ml LB + ab media
2. Incubate culture overnight at 37C
3. Transfer culture to 2 ml microfuge tube
4. Spin cells down at 13K rpm for 2 min at RT and remove supernatant
5. Resuspend cells in 250 ul Buffer P1 (stored at 4 C)
6. Add 250 ul Buffer P2 and mix thoroughly by inverting-- the solution should turn blue
7. Add 350 ul Buffer N3 and mix immediately and thoroughly by inverting-- the solution should turn colorless
8. Centrifuge sample at 13K rpm for 10 minutes
9. Transfer supernatant to a fresh QIAprep spin column, leaving cell debris pellet behind
10. Centrifugre supernatant into column at 13K rpm for 1 minute
11. Remove the flowthrough
12. Wash column with 0.5 ml Buffer PB; apply to column and spin through at 13K rpm for 1 minute
13. Remove the flowthrough
14. Wash column with 0.75 ml Buffer PE; apply to column and spin through at 13K rpm for 1 minute
15. Remove the flowthrough
16. Spin out residual liquid at 13K rpm for 1 minute
17. Place column in a fresh 1.5 ml microfuge tube
18. Elute DNA; apply 40 ul Buffer EB to column, incubate at room temperature for 2 minutes and spin out of column at 13K rpm for one minute