



## Quick Protocol Reference Card Spin Doctor MB Mini Prep Kit

### Protocol

1. Grow 3ml of culture for plasmids and 5ml for large/low copy constructs
2. Spin down pellet for 5 minutes at 12,000 RPM
3. HIGHLY RECOMMENDED TO GENERATE MAX YIELDS (especially for **BACs, low copy, and large constructs**) Optional: Pipette additional 1.4mL of culture into the tube containing the pellet and spin down for 5 minutes at 12,000 RPM – pour off supernatant – your pellet will now be the result of 2.8mL of culture
4. Re-suspend pellet in 200uL of Solution 1 and vortex
5. Add Solution 2 and mix by gently inverting
6. Add Solution 3 and mix by gently inverting
7. Spin down pellet for 10 minutes
8. Pipette off 400uL of supernatant into a clean 1.7mL tube
9. Add Isopropanol - mix by inverting 20 times and spin down pellet at maximum speed for 10 minutes in a microcentrifuge
10. Pour off supernatant and pipette off remains - allow to air dry for 5 minutes
11. Re-suspend one RNase tube using 1mL of Solution 4 - add 50uL of the resuspended RNase to each prep and pipette briefly to resuspend the RNA/DNA pellet
12. Incubate at 37°C for 15 minutes - at 7 ½ minutes mix by inverting
13. Add Solution 5 and mix by inverting
14. Add 100uL of Isopropanol and invert 20 times - spin down pellet for 10 minutes at maximum speed in a microcentrifuge - pour off supernatant.
15. Add 70% Ethanol – invert 10 times and microfuge at full speed for 2 min.
16. Repeat step 14 (Ethanol wash)
17. Pipette off remaining ethanol and air dry for 5 minutes
18. Re-suspend the DNA in 75uL of 10mM Tris Buffer pH 8.0 – incubate at 57°C for 10 minutes



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