

**Protocol for Using Wizard Plus SV Minipreps Plasmid Purification Kit**  
**Chris Song, 7-29-05**

1. pellet **4ml** overnight culture in 2ml microfuge tubes for 1 min.
2. thoroughly resuspend pellet with **250ul** cell resuspension solution.
3. add **250ul** cell lysis solution; invert 4 times to mix well.
4. add **10ul** alkaline protease solution; invert 4 times to mix well.
5. add **350ul** neutralization solution; invert 4 times to mix well.
6. centrifuge at top speed for **10** minutes at room temperature (Note: it is very important to centrifuge well).
7. prepare a tube of sterile ddH<sub>2</sub>O (2ml tube), put it into 70 C incubator.
8. insert spin column into collection tube and decant cleared lysate into spin column.
9. centrifuge at top speed for **1min** at room temperature. Throw away the liquid.
10. add **500ul** wash solution (ethanol added), centrifuge at top speed for **20s** at room temperature. Throw away the liquid.
11. add **500ul** wash solution (ethanol added), centrifuge at top speed for **1.5min** at room temperature. Discard collection tube with flowthrough. If the spin column still has any of the wash solution with it, centrifuge again for 1min at top speed.
12. Transfer spin column to a 1.7ml centrifuge tube. Add 100ul of pre-heated dd water to the spin column; spin it at top speed for 1min.