

Wizard® Plus SV Minipreps DNA Purification System

INSTRUCTIONS FOR USE OF PRODUCTS A1330, A1340, A1460, A1465 AND A1470.

Quick
PROTOCOL

Centrifugation Protocol

Production of Cleared Lysate

1. Pellet 1–10ml of overnight culture for 5 minutes.
2. Thoroughly resuspend pellet with 250µl of Cell Resuspension Solution.
3. Add 250µl of Cell Lysis Solution to each sample; invert 4 times to mix.
4. Add 10µl of Alkaline Protease Solution; invert 4 times to mix. Incubate 5 minutes at room temperature.
5. Add 350µl of Neutralization Solution; invert 4 times to mix.
6. Centrifuge at top speed for 10 minutes at room temperature.

Binding of Plasmid DNA

7. Insert Spin Column into Collection Tube.
8. Decant cleared lysate into Spin Column.
9. Centrifuge at top speed for 1 minute at room temperature. Discard flowthrough, and reinsert Column into Collection Tube.

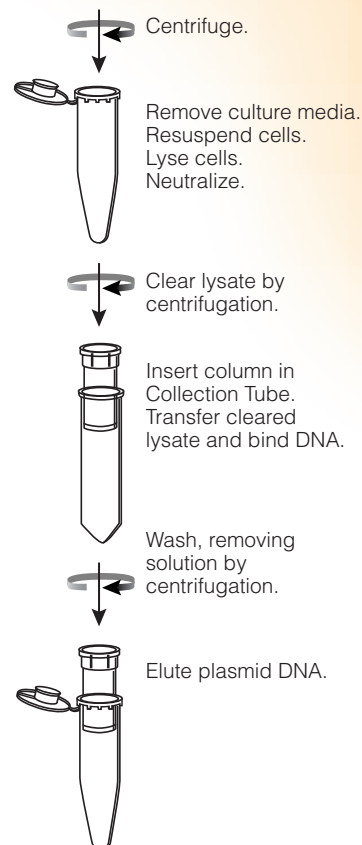
Washing

10. Add 750µl of Wash Solution (ethanol added). Centrifuge at top speed for 1 minute. Discard flowthrough and reinsert column into Collection Tube.
11. Repeat Step 10 with 250µl of Wash Solution.
12. Centrifuge at top speed for 2 minutes at room temperature.

Elution

13. Transfer Spin Column to a sterile 1.5ml microcentrifuge tube, being careful not to transfer any of the Column Wash Solution with the Spin Column. If the Spin Column has Column Wash Solution associated with it, centrifuge again for 1 minute at top speed, then transfer the Spin Column to a new, sterile 1.5ml microcentrifuge tube.
14. Add 100µl of Nuclease-Free Water to the Spin Column. Centrifuge at top speed for 1 minute at room temperature.
15. Discard column, and store DNA at –20°C or below.

Overnight culture



1581MB09_9A

Additional protocol information is available in Technical Bulletin #TB225, available online at: www.promega.com

ORDERING/TECHNICAL INFORMATION:

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INSTRUCTIONS FOR USE OF PRODUCTS A1330, A1340, A1460, A1465 AND A1470.

Quick
PROTOCOL

Vacuum Protocol

Production of Cleared Lysate

1. Pellet 1–10ml of overnight culture for 5 minutes.
2. Thoroughly resuspend pellet with 250µl of Cell Resuspension Solution.
3. Add 250µl of Cell Lysis Solution to each sample; invert 4 times to mix.
4. Add 10µl of Alkaline Protease Solution; invert 4 times to mix. Incubate 5 minutes at room temperature.
5. Add 350µl of Neutralization Solution; invert 4 times to mix.
6. Centrifuge at top speed for 10 minutes at room temperature.

Binding of Plasmid DNA

7. Attach Vacuum Adapter to manifold port, and insert Spin Column into Adapter.
8. Decant cleared lysate into column.
9. Apply vacuum to pull liquid through column. Release vacuum when all liquid has passed through column.

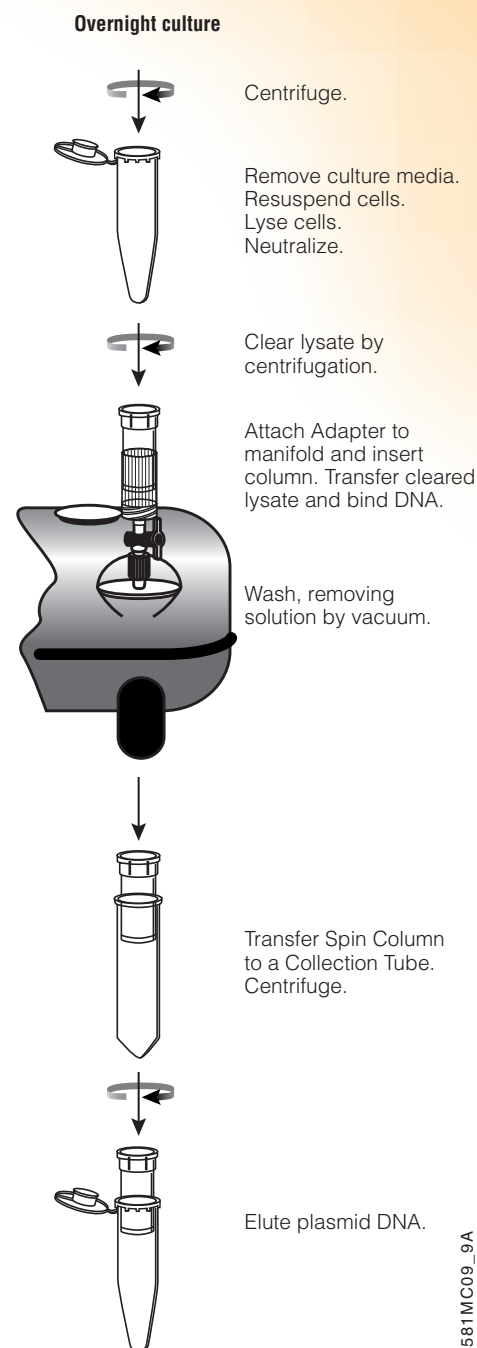
Washing

10. Add 750µl of Wash Solution (ethanol added). Apply vacuum to pull solution through column.
11. Turn off vacuum, and repeat Step 10 with 250µl of Wash Solution.
12. Dry by applying a vacuum for 10 minutes.
13. Transfer the column to a 2ml Collection Tube, and centrifuge at top speed for 2 minutes.

Elution

14. Transfer column to a sterile 1.5ml microcentrifuge tube.
15. Add 100µl of Nuclease-Free Water to the column. Centrifuge at top speed for 1 minute at room temperature.
16. Discard column. Store DNA at –20°C or below.

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