

PROMEGA Wizard® SV Gel and PCR Clean-up System

1. Gel slice and PCR product preparation

a) Dissolving the gel slice:

- Following electrophoresis, excise DNA band from gel and place gel slice in a 1.5 mL microcentrifuge tube.
- Add 10 μ L Membrane Binding Solution per 10 mg gel slice. Vortex and incubate at 50-65°C until the gel slice is completely dissolved.

b) Processing PCR amplifications:

- Add an equal volume of Membrane Binding Solution to the PCR amplification.

2. Binding of DNA

- Insert SV Minicolumn into Collection Tube
- Transfer dissolved gel mixture or prepared PCR product to the Minicolumn assembly. Incubate at room temperature for 1 minute.
- Centrifuge at 16,000 x g for 1 minute. Discard flowthrough and reinsert Minicolumn into Collection Tube

3. Washing

- Add 700 μ L Membrane Wash Solution (ethanol added). Centrifuge at 16,000 x g for 1 minute. Discard flowthrough and reinsert Minicolumn into Collection Tube.
- Repeat the last step with 500 μ L Membrane Wash Solution. Centrifuge at 16,000 x g for 5 minutes.
- Empty the Collection Tube and recentrifuge the column assembly with the microcentrifuge lid open (or off) to allow evaporation of any residual ethanol.

4. Elution

- Carefully transfer Minicolumn to a clean 1.5 mL microcentrifuge tube.
- Add 50 μ L of Nuclease-Free Water to the Minicolumn. Incubate at room temperature for 1 minute. Centrifuge at 16,000 x g for 1 minute.
- Discard Minicolumn and store DNA at 4°C or -20°C.

From: iGEM Bielefeld-CeBiTec