PCR Purification (Promega Wizard SV Gel and PCR Clean-Up)

Notes: Prewarm elution buffer to 65 °C

Dissolving the Gel Slice

1. Following electrophoresis, excise DNA band from gel and place gel slice in a

   1.5ml microcentrifuge tube.

2. Add 10µl Membrane Binding Solution per 10mg of gel slice. Vortex and incubate at 50–65°C until gel slice is completely dissolved.

   Processing PCR Amplifications

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

   Binding of DNA

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

   Washing

7. Add 700µl Membrane Wash Solution (ethanol added). Centrifuge at 16,000 × g for 1 minute. Discard flowthrough and reinsert Minicolumn into Collection Tube.
8. Repeat Step 4 with 500µl Membrane Wash Solution. Centrifuge at 16,000 × g for 5 minutes
9. Empty the Collection Tube and recentrifuge the column assembly for 1 minute with the microcentrifuge lid open (or off) to allow evaporation of any residual ethanol.

   Elution

10. Carefully transfer Minicolumn to a clean 1.5ml microcentrifuge tube
11. Add 50µl of Nuclease-Free Water to the Minicolumn. Incubate at room temperature for 1 minute. Centrifuge at 16,000 × g for 1 minute.