DNA Purification Through Gel Extraction

During a gel extraction, the majority of the DNA is recovered in the first 20uL of the elution. This protocol provides a more concentrated DNA solution than is produced by the recommended method. This concentrated DNA solution can be used directly in ligation reactions, allowing for the bypass of precipitation procedures normally performed after gel extractions.

1) Excise desired DNA fragment from a 1% agarose and place in a 1.5mL microfuge tube.

2) Add 500uL of membrane binding solution to the microfuge tube and incubate with shaking at 65°C until all gel is dissolved.

3) Place an SV minicolumn into a collection tube and add the dissolved gel solution to the SV minicolumn.

4) Incubate solution in the SV minicolumn for 1 min at room temperature and then centrifuge at 16 000xg for 1min.

5) Discard flow through and add 700uL of membrane wash solution to the SV minicolumn and centrifuge at 16 000xg for 1 min.

6) Discard flow through and add 500uL of membrane wash solution to the SV minicolumn and centrifuge at 16 000xg for 1 min.

7) Discard flow through and centrifuge again at 16 000xg for 5 min.

8) Discard flow through and centrifuge again at 16 000xg for 1 min to remove any residual ethanol.

9) Transfer SV minicolumn to a clean 1.5mL microfuge tube and then gently add 20uL of nuclease free water to the center of the SV minicolumn filter and let stand at room temperature for 1 min then centrifuge at 16 000xg for 1 min.

10) Discard SV minicolumn and store DNA elution (15uL) at -20°C.