Gel Extraction

Material

E.Z.N.A.® Gel Extraction Kit (OMEGA Code No.D2500-01)

Procedure

1. Perform agarose gel/ethidium bromide electrophoresis to fractionate DNA fragments.
2. When adequate separation of bands has occurred, carefully excise the DNA fragment of interest using a wide, clean, sharp scalpel.
3. Determine the appropriate volume of the gel slice by weighing it in a clean 1.5 Ml EP tube. Assuming a density of 1 g/mL, the volume of gel is derived as follows: a gel slice of mass 0.3 g will have a volume of 0.3 mL.
4. Add 1 volume Binding Buffer (XP2).
5. Incubate at 60°C for 7 minutes or until the gel has completely melted. Vortex or shake the tube every 2-3 minutes.
6. Insert a HiBind® DNA Mini Column in a 2 mL Collection Tube.
7. Add no more than 700 µL DNA/agarose solution from Step 5 to the HiBind® DNA Mini Column.
8. Centrifuge at 10,000 x g for 1 minute at room temperature.
9. Discard the filtrate and reuse collection tube.
10. Repeat Steps 7-9 until all of the sample has been transferred to the column.
11. Add 300 µL Binding Buffer (XP2).
12. Centrifuge at maximum speed (≥13,000 x g) for 1 minute at room temperature.
13. Discard the filtrate and reuse collection tube.
14. Add 700 µL SPW Wash Buffer.
15. Centrifuge at maximum speed for 1 minute at room temperature.
16. Discard the filtrate and reuse collection tube.
17. Centrifuge the empty HiBind® DNA Mini Column for 2 minutes at maximum speed to dry the column matrix.
18. Transfer the HiBind® DNA Mini Column to a clean 1.5 mL EP tube.
19. Add 30-50 µL Elution Buffer or deionized water directly to the center of the column membrane.
20. Let sit at room temperature for 2 minutes.
21. Centrifuge at maximum speed for 1 minute.
22. Store DNA at -20°C.