Gel Extraction

Materials

- Agarose gel
- QIAquick Gel Extraction Kit

Procedure

1. Excise the DNA fragment from the agarose gel with a clean, sharp scalpel
2. Weigh the gel slice in a colorless tube. Add 3 volumes Buffer QG to 1 volume gel (100 mg = 100µL). For >2% agarose gels, add 6 volumes Buffer QG
3. Incubate at 50ºC for 10 minutes (or until the gel slice has completely dissolved). Vortex the tube every 2-3 minutes to help dissolve the gel
4. After the gel slice has dissolved completely, check that the color of the mixture is yellow (similar to Buffer QG without dissolved agarose). If the color of the mixture is orange or violet, add 10µL 3M sodium acetate, pH5.0, and mix. The color of the mixture will turn yellow
5. Add 1 gel volume of isopropanol to the sample and mix
6. Place a QIAquick spin column in a provided 2 mL collection tube
7. To bind DNA, apply the sample to QIAquick column and centrifuge for 1 min. Discard the flow-through and place the QIAquick column back into the same tube. For sample volumes of >800 µL, load and spin again
8. To wash, add 0.75 mL Buffer PE to QIAquick column and centrifuge for 1 min. Discard the flow-through and place the QIAquick column back into the same tube
9. Centrifuge the QIAquick column once more in the provided 2 ML collection tube for 1 min at 13'000 rpm to remove residual buffer
10. Place QIAquick column into a clean 1.5 mL microcentrifuge tube
11. To elute DNA, add 50 µL Buffer EB or water to the center of the QIAquick membrane and centrifuge the column for 1 min. For increased DNA concentration, add 30 µL Buffer EB to the center of the QIAquick membrane, let the column stand for 1 min, and then centrifuge for 1 min. After the addition of Buffer EB to the QIAquick membrane, increasing the incubation time to up to 4 min can increase the yield of purified DNA