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Gel DNA Extraction Protocol

- The precipitates that appear during storage, especially at low temperature, in some buffers are easily dissolved by incubating the bottles at 60°C.
- Add 40ml absolute ethanol to Wash Buffer II.
- Set thermoblock or water bath at 50-60°C.
- Preheat Elution Buffer at this temperature.

1- Excise the gel slice containing the DNA band with a clean and sharp scalpel.

2- Add 2-3 volumes of **Lysis Buffer** to 1 volume of gel and incubate at 50-60°C for 10 min (until the gel completely dissolved). During this period, you can vortex several times.

3- Add 100µl (same volume of gel) isopropanol then vortex and spin briefly.

4- Transfer the mixture into the spin column placed in a collection tube. Centrifuge for 1 min at 10000 rpm.

5- Discard flow-through and place the column in a new collection tube.

6- Add 500µl **Wash Buffer II** to each tube.

7- Centrifuge for 1 min at 10000 rpm. Then proceed centrifugation at full speed (14000 rpm) for 2-3 min to dry the membrane completely.

8- Place the column in a clean 1.5ml microtube. Open the lid of the column and apply 20-50µl preheated **Elution Buffer** to the center of the membrane.

9- Close the lid and incubate at 50-60°C for 2-5 min. Centrifuge at full speed (>12000 rpm) for 1 min.

10- Store eluted DNA at -20°C.