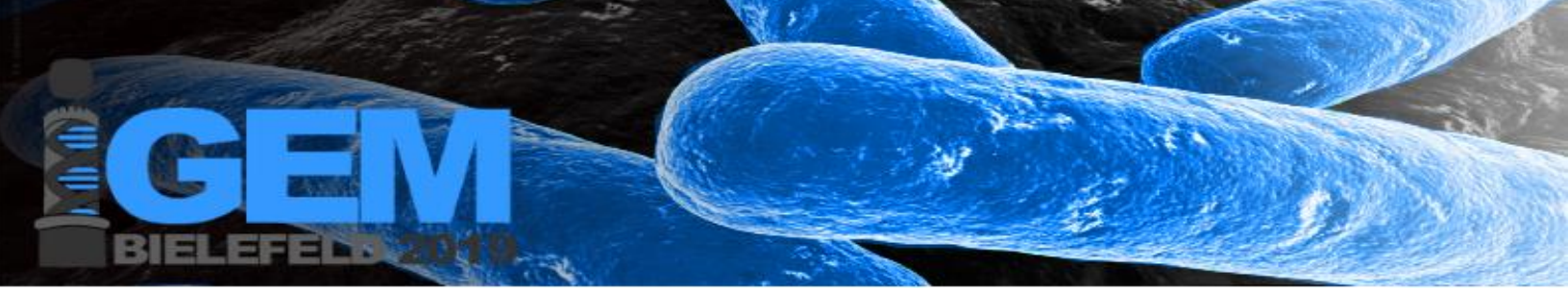

Macherey Nagel NucleoSpin® Gel and PCR Clean-up

1. **Preparation of the sample**
   a) DNA extraction from agarose gels:
      - Add 200 µL Buffer NT1 for each 100 mg of agarose gel < 2%
      - Incubate sample for 5-10 min at 50°C. Vortex the sample briefly every 2-3 min until the gel slice is completely dissolved.

   b) PCR clean-up:
      - Mix 1 volume of sample with 2 volumes of buffer NT1 (e.g., mix 100 µL PCR reaction and 200 µL Buffer NT1)

2. **Bind DNA**
   Place a NucleoSpin® Gel and PCR Clean/up column into a collection tube (2 mL) and load up to 700 µL sample. Centrifuge for 30 s at 11,000 x g. Discard flow/through and place the column back into the collection tube.

   Load remaining sample if necessary and repeat the centrifugation step.

3. **Wash silica membrane**
   Add 700µL Buffer NT3 to the NucleoSpin® Gel and PCR Clean/up column. Centrifuge for 30 s at 11,000 x g. Discard flow/through and place the column back into the collection tube.

   Repeat this washing step to minimiye chaotropic salt carrz/over and improve A_{260}/A_{230} values.

4. **Dry silica membrane**
   Centrifuge for 1 min at 11,000 x g to remove Buffer NT3 completely. Make sure the spin column does not come in contact with the flow-through while removing it from the centrifuge and the collection tube.

5. **Elute DNA**
   Place the NucleoSpin® Gel and PCR Clean/up column into a new 1.5 mL microcentrifuge tube (not provided). Add 15-30 µL Buffer NE and incubate at room temperature (18-25°C) for 1 min. Centrifuge for 1 min at 11,000 x g.

From: iGEM Bielefeld-CeBiTec

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