Quick-Start Protocol April 2016

MinElute® PCR Purification Kit

The MinElute PCR Purification Kit (cat. nos. 28004 and 28006) can be stored at room temperature (15–25°C) for up to 12 months if not otherwise stated on label. Store spin columns at 2–8°C upon arrival.

Further information

- *MinElute Handbook*: **www.qiagen.com/HB-2069**
- Safety Data Sheets: **www.qiagen.com/safety**
- Technical assistance: **support.qiagen.com**

Notes before starting

- This protocol is for cleanup of up to 5 µg PCR product (70 bp to 4 kb).
- Add ethanol (96–100%) to Buffer PE concentrate before use (see bottle label for volume).
- All centrifugation steps are carried out at 17,900 x *g* (13,000 rpm) in a conventional tabletop microcentrifuge at room temperature (15–25°C).
- Add 1:250 volume pH indicator I to Buffer PB. Add pH indicator I to the entire buffer contents. Do not add pH indicator I to buffer aliquots. The yellow color of Buffer PB with pH indicator I indicates a pH of ≤7.5. The adsorption of DNA to the membrane is efficient only at pH ≤7.5.

  Note: If the purified PCR product is to be used in sensitive microarray applications, it may be beneficial to use Buffer PB without addition of pH indicator I.
- Symbols: ● centrifuge processing; ▲ vacuum processing.

1. Add 5 volumes of Buffer PB to 1 volume of the PCR reaction and mix. Check that the color of the mixture is yellow (similar to Buffer PB without the PCR sample). If the color of the mixture is orange or violet, add 10 µl 3 M sodium acetate, pH 5.0, and mix. The color of the mixture will turn to yellow.
2. Place a MinElute column ● in a provided 2 ml collection tube or ▲ into a vacuum manifold. See the *MinElute Handbook* for details on how to set up a vacuum manifold.
3. Apply the sample to the MinElute column and ● centrifuge for 1 min or ▲ apply vacuum until the entire sample has passed through the column. ● Discard flow-through and place the MinElute column back into the same collection tube.
4. Add 750 µl Buffer PE to the MinElute column and ● centrifuge for 1 min or ▲ apply vacuum. ● Discard flow-through and place the MinElute column back in the same collection tube.
5. Centrifuge the column in a 2 ml collection tube (provided) for 1 min. Residual ethanol from Buffer PE will not be completely removed unless the flow-through is discarded before this additional centrifugation.
6. Place each MinElute column in a clean 1.5 ml microcentrifuge tube.
7. To elute DNA, add 10 µl Buffer EB (10 mM Tris·Cl, pH 8.5) or water to the center of the MinElute membrane. (Ensure that the elution buffer is dispensed directly onto the center of the membrane for complete elution of bound DNA.) Let the column stand for 1 min, and then centrifuge the column for 1 min.
8. If the purified DNA is to be analyzed on a gel, add 1 volume of Loading Dye to 5 volumes of purified DNA. Mix the solution by pipetting up and down before loading the gel.

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For up-to-date licensing information and product-specific disclaimers, see the respective QIAGEN kit handbook or user manual.