Quick-Start Protocol March 2016

QIAquick® Nucleotide Removal Kit

The QIAquick Nucleotide Removal Kit (cat. nos. 28304 and 28306) can be stored at room temperature (15–25°C) for up to 12 months if not otherwise stated on label.

Further information

- *QIAquick Spin Handbook*: **www.qiagen.com/HB-1196**
- Safety Data Sheets: **www.qiagen.com/safety**
- Technical assistance: **support.qiagen.com**

Notes before starting

- This protocol is for cleanup of up to 10 µg oligonucleotides (17–40mers) and/or DNA fragments (40 bp to 10 kb) from enzymatic reactions.
- Add isopropanol to Buffer PNI before use (see bottle label for volume).
- Add ethanol (96–100%) to Buffer PE concentrate before use (see bottle label for volume).
- All centrifugation steps are carried out in a conventional tabletop microcentrifuge at room temperature (15–25°C).

1. Add 10 volumes of Buffer PNI to 1 volume of sample and mix. For DNA fragments ≥100 bp, only 5 volumes of Buffer PNI are required.
2. Place a QIAquick spin column in a provided 2 ml collection tube.
3. Apply the sample to the QIAquick spin column and centrifuge for 1 min at 6000 rpm.
4. **For radioactive samples**: Discard the collection tube containing radioactive flow-through appropriately and place the QIAquick spin column into a clean 2 ml collection tube.

   For non-radioactive samples: Discard the flow-through and place the QIAquick spin column back into the same collection tube.

5. **For radioactive samples**: Add 500 µl Buffer PE and centrifuge for 1 min at 3800 x *g* (6000 rpm). Discard the flow-through appropriately and repeat the wash with another 500 µl Buffer PE.

   For non-radioactive samples: Add 750 µl Buffer PE and centrifuge for 1 min at 3800 x *g* (6000 rpm).
6. Discard the flow-through and place the QIAquick spin column back in the same collection tube. Centrifuge for an additional 1 min at 17,900 x *g* (13,000 rpm). (Residual ethanol from Buffer PE will not be completely removed unless the flow-through is discarded before this additional centrifugation.)
7. Place the QIAquick spin column in a clean 1.5 ml microcentrifuge tube.
8. To elute DNA, add 100–200 µl Buffer EB (10 mM Tris·Cl, pH 8.5) or water to the center of the QIAquick membrane and centrifuge for 1 min at 17,900 x *g* (13,000 rpm). Alternatively, for increased DNA concentration, add 30–50 µl elution buffer to the center of the QIAquick membrane, let the column stand for 1 min and then centrifuge.
9. 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.

Scan QR code for handbook.

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

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