Quick-Start Protocol April 2016

# DyeEx® 96 Kit

The DyeEx 96 Kit (cat. nos. 63181 and 63183) should be stored at 2–8°C for up to 12 months if not otherwise stated on label. Do not freeze.

## Further information

- *DyeEx Handbook*: **www.qiagen.com/HB-0582**
- Safety Data Sheets: **www.qiagen.com/safety**
- Technical assistance: **support.qiagen.com**

## Notes before starting

- A multichannel pipet facilitates handling of sequencing samples.
- The use of DyeEx 96 plates requires a suitable centrifuge capable of centrifuging microplates of 4.5 cm total height. All centrifugation steps are carried out at 1000 x *g* at room temperature (15–25°C). The appropriate speed can be calculated:

  rpm = 1000 x $\sqrt{1000/1.12\ r}$ (r = radius of rotor in mm).
- After centrifugation, the gel-bed surface in the wells of the DyeEx 96 plate may vary due to the differing centrifugal force in the different wells. This is normal and has no effect on the performance of the DyeEx 96 procedure.
- Always use the waste collection plates provided with the DyeEx 96 Kit.

1. Take the DyeEx 96 plate out of the bag, and remove the tape sheets first from the bottom and then from the top of the DyeEx 96 plate. When handling the DyeEx 96 plate, ensure that it remains horizontal.
2. Place the DyeEx 96 plate on the top of the collection plate (provided; reusable) and centrifuge for 1 min at the calculated speed. Discard the flow-through. Always use the waste collection plates provided with the DyeEx 96 Kit.

3. Place the DyeEx 96 plate on top of the collection plate, add 300 µl deionized water to each well and centrifuge for 3 min at the calculated speed.
4. Carefully place the DyeEx 96 plate on an appropriate elution plate with a suitable adapter.

   **Note**: For direct loading onto sequencer, place the DyeEx 96 plate on a 96-well PCR plate or on 12 x 8-well strips, in each case with an appropriate adapter. Suitable 96-well PCR plates include those from ABgene (ThermoFast 96 PCR Plates, VWR cat. no. 732-4828), and suitable adapters include those from PE Biosystems (MicroAmp® Base, cat. no. N801-0531).

   The DyeEx 96 plate sits securely in the centrifuge rotor when the tops of the elution-plate wells are in direct contact with the base of the DyeEx 96 plate.
5. Slowly apply the sequencing samples in a volume of 10–20 µl to the gel bed of each well.

   **Note**: Pipet the sequencing reaction directly onto the center of the gel-bed surface, without touching the reaction mixture or the pipet tip to the sides of the wells. The samples should be pipetted slowly so that they are absorbed into the gel and do not flow down the sides of the gel bed. Avoid touching the gel-bed surface with the pipet tip.
6. Centrifuge for 3 min at the calculated speed. The eluate contains the purified sequencing reaction.

   For most sequencers, it is possible to load the eluate directly onto the sequencer.

   **Optional**: If using a formamide loading buffer, dry the samples and proceed according to the instructions provided with the DNA sequencer.

Scan QR code for handbook.

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