

Quick-Start Protocol

EZ1&2® ccfDNA Kit

The EZ1&2 ccfDNA Kit (cat. no. 954854) is shipped at room temperature (15–25°C). Upon receipt, store all kit components dry at room temperature.

Further information

- EZ1&2 ccfDNA Kit Handbook: www.qiagen.com/HB-2915
- Safety Data Sheets: www.qiagen.com/safety
- Technical assistance: support.qiagen.com

Notes before starting

- Plasma samples should be at room temperature before use. If plasma/serum was stored frozen, thaw the samples by incubating at 37°C in a water bath.
- If the sample volume is less than 2 mL, 4 mL, 8 mL, or 10 mL (only on the EZ2 Connect), adjust the volume with PBS to 2 mL, 4 mL, 8 mL, or 10 mL.

Procedure

- 1. Load reagent cartridges into the cartridge rack.
- 2. Vortex the Magnetic Bead Suspension EZ briefly in an upright position.
- 3. Turn the tube with the cap facing downwards and vortex again to ensure a homogeneous bead suspension.
- 4. Briefly centrifuge to collect all the liquid at the bottom of the tube. Remove the cap from the tube with the bead solution and place in position 11 of the EZ1&2 ccfDNA cartridge.

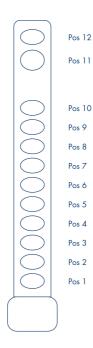


Figure 1. EZ1&2 ccfDNA cartridge.

- 5. Transfer 900 µL Elution Buffer EZE into position 12 of the EZ1&2 ccfDNA cartridge.
- 6. Remove caps of all tubes and prepare the Tip Rack as follows (see Figure 2):
 - Position 4/A: Tip holder with Filter Tip (provided)
 - Position 3/B: new large volume tube (7 mL) (provided)
 - Position 2/C: new large volume tube (7 mL) (provided)
 - Position 1/D: new 1.5 mL elution tube (provided)



Figure 2. Tip Rack - Large Volume.

- 7. For 2 mL/4 mL protocol: Transfer 2 mL/4 mL sample into large volume tube in position 2/C. Place an empty large volume tube in position 3/B.
 - For 8 mL protocol: Split your sample and transfer 4 mL into each of the large volume tubes in positions 2/C and 3/B.
 - For 10 mL protocol (only for EZ2 Connect): Split your sample and transfer 5 mL into each of the 7 mL tubes in positions 2/C and 3/B.

Procedure on the EZ2® Connect

- 8. Turn on the EZ2 Connect instrument.
- Tap DNA on the Applications panel and select the "EZ1&2 ccfDNA Kit" and select Next.
 Follow onscreen instructions for selection of protocol, parameter definition, sample position selection, sample IDs, and worktable setup.
- 10. Open the instrument door. Load the cartridge rack into the instrument.
- 11. Place the tip rack into the instrument.
- 12. Close the instrument door; select **Start** to initiate the EZ1&2 ccfDNA protocol.
- 13. The display will show "Protocol finished" when the run is completed. Select **Finish**.

 Open the instrument hood. Remove the elution tubes containing the purified ccfDNA from position 1/D of the tip rack. Discard the sample preparation waste (in tubes in positions 2/C and 3/B and, in the case of the 8 mL and 10 mL protocols, in position 4/A) (see Figure 2).
 - **Optional**: Follow onscreen instructions for UV decontamination of worktable surfaces.
- 14. Perform regular maintenance after each run. Select **Finish** to return to the Home Screen.

Procedure on the EZ1® Advanced XL

- 1. For use on EZ1 Advanced XL instrument, insert the EZ1 Advanced XL ccfDNA Card completely into the card slot and turn on the instrument. Please make sure the instrument is turned off prior to inserting the card.
- Select Start to initiate the EZ1&2 ccfDNA protocol. Follow on-screen instructions for selection of protocol, data tracking, and worktable setup. Close the instrument door; select Start to start the protocol.
- 3. Open instrument door. Load the cartridge rack into the instrument.
- 4. Place the tip rack into the instrument.
- 5. The display will show "Protocol finished" when finished. Press the Esc key.
- 6. Open the instrument door. Remove the elution tubes containing the purified ccfDNA (in 75 µL) from position 1 of the tip rack. Discard the sample preparation waste (in tubes in positions 2 and 3 and, in the case of the 8 mL protocol, in position 4. Press the Enter key.

The report file is transferred automatically.

Optional: Follow onscreen instructions for UV decontamination of worktable surfaces.

7. Perform regular maintenance after each run. Press the Esc key to return to the Main Menu.

Document Revision History

Date	Description
11/2021	Initial release
08/2023	Incorporated additional protocol from the revision 3 of the Handbook.
11/2025	Added the 10 mL protocol for the EZ2 Connect.



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