

EpiTect[®] Plus LyseAll Lysis Kit

The EpiTect Plus LyseAll Lysis Kit included in the EpiTect Plus LyseAll Bisulfite Kit (cat. no. 59164) can be stored at room temperature (15–25°C) for up to 6 months.

For more information, please refer to the *EpiTect Plus Bisulfite Conversion Handbook*, which can be found at www.qiagen.com/handbooks.

For technical assistance, please call toll-free 00800-22-44-6000, or find regional phone numbers at www.qiagen.com/contact.

Notes before starting

- Prepare reagents as described in EpiTect Plus DNA Bisulfite Kit (included).
- Perform all centrifugation steps at room temperature (15–25°C).
- Do not place samples on ice after beginning lysis procedure.

Lysis of whole blood samples

1. Dilute blood samples (maximum 20 μ l) 1:20 with Buffer EL.
2. Incubate at room temperature (15–25°C) for 10–15 min, inverting the tubes several times during incubation.
3. Centrifuge at maximum speed for 5 min. Discard supernatant and add an additional 125 μ l Buffer EL.
4. Centrifuge at maximum speed for 1 min. Discard supernatant.
5. Resuspend cells in 10 μ l PBS and transfer into a 200 μ l reaction tube or into 8-well strips (not provided).
6. Add 10 μ l distilled water, 15 μ l Lysis Buffer FTB, and 5 μ l proteinase K. Vortex and centrifuge briefly. Proceed to step 12.

Lysis of cultured cells

7. Harvest cells according to your current protocol.
8. Resuspend cells in 10 μl PBS in a 200 μl reaction tube or into 8-well strips (not provided). Do not use more than 1×10^5 cells per 10 μl PBS.
9. Add 10 μl distilled water, 15 μl Lysis Buffer FTB, and 5 μl proteinase K. Vortex and centrifuge briefly. Proceed to step 12.

Lysis of tissue samples

10. Place the tissue sample ($\leq 100 \mu\text{g}$) in a 200 μl reaction tube or 8-well strip and add 20 μl distilled water.
11. Add 15 μl Lysis Buffer FTB and 5 μl proteinase K. Vortex and centrifuge briefly.
12. Incubate samples for 30 min at 56°C.
13. Add 800 μl RNase-free water to each aliquot of Bisulfite Mix needed, and vortex until Bisulfite Mix is completely dissolved. This may take up to 5 min. Dissolving the Bisulfite Mix may require heating the solution to 60°C.
14. Set up the bisulfite reactions in 200 μl PCR tubes (not provided) according to Table 1. Add each component in the order listed.
15. To perform the bisulfite DNA conversion, proceed to Protocol 1 “Bisulfite conversion of DNA” included in the supplied EpiTect Plus DNA Bisulfite Kit.

Table 1. Bisulfite reaction setup

Component	Volume per reaction (μl)
Lysis reaction	40
Bisulfite Mix (dissolved), see step 13	85
DNA Protect Buffer	15
Total volume	140

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

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