

User-Developed Protocol:

Isolation of genomic DNA from blood (erythrocyte lysis) using the QIAGEN® Genomic-tip 100/G

This procedure has been adapted by customers from the Genomic-tip Protocols, and is for use with the QIAGEN® Genomic-tip. **It has not been thoroughly tested or optimized by QIAGEN.**

This protocol is designed for genomic DNA quantities of up to 100 µg from 5.0 ml of human whole blood. The size of yielded chromosomal DNA ranges from 20–150 kb. Genomic DNA prepared by this method is generally suitable for restriction digestions, Southern blotting, and PCR. The following protocol is carried out in 50 ml screw-cap tubes.

Please be sure to read the *QIAGEN Genomic DNA Handbook* and the detailed Sample Preparation and Lysis Protocol for Blood carefully before beginning this procedure.

Important note before starting

- If using QIAGEN Protease in step 6, follow the instructions given on the label or product sheet for reconstitution before use.

Procedure

1. **To 5.0 ml whole blood add 5 µl of 1% Saponin solution and mix by inverting the tube 3–5 times until the solution is clear.**
Note: Frozen blood may not appear clear after lysis.
2. **Centrifuge at 1500 rpm for 20 min at room temperature and discard the supernatant.**
3. **Add 15.0 ml PBS-buffer, pH 7.4, and vortex for 5–15 s to resuspend and wash the leukocyte pellet.**
4. **Centrifuge again at 1500 rpm for 15 min at room temperature and discard the PBS wash solution.**
5. **Add 5 ml of ice-cold Buffer G2 and resuspend the pellet by vortexing. Ensure that it is thoroughly resuspended.**
6. **Add 200 µl QIAGEN Protease or Proteinase K solution.**
7. **Incubate at 50°C for 2 h.**
8. **Follow the QIAGEN Genomic-tip 100/G procedure of the Sample Preparation and Lysis Protocol for Blood in the *QIAGEN Genomic DNA Handbook*.**

QIAGEN handbooks can be requested from QIAGEN Technical Service or your local QIAGEN distributor. Selected handbooks can be downloaded from www.qiagen.com/literature/handbooks/default.asp. Material safety data sheets (MSDS) for any QIAGEN product can be downloaded from www.qiagen.com/ts/msds.asp.

Trademark: QIAGEN® (QIAGEN). The PCR process is covered by U.S. Patents 4,683,195 and 4,683,202 and foreign equivalents owned by Hoffmann-La Roche AG. © 2001 QIAGEN, all rights reserved.