
# User-Developed Protocol:

# Isolation of genomic DNA from sperm using the QIAamp® DNA Mini Kit; protocol 2

This procedure has been adapted by customers from the QIAamp® Tissue Protocol, and is for use with the QIAamp DNA Mini Kit. **It has not been thoroughly tested and optimized by QIAGEN.**

Please note that lysis time will vary depending on the size and density of the source material.

Please be sure to read the QIAGEN® *QIAamp DNA Mini Kit and QIAamp DNA Blood Mini Kit Handbook* and the detailed QIAamp Tissue Protocol carefully before beginning this procedure.

## Procedure

1. **Place 100 µl sperm in a 1.5 ml microcentrifuge tube and add 100 µl Buffer X2. Incubate at 55°C until the sample is dissolved (at least 1 h). Invert the tube occasionally to disperse the sample, or place on a rocking platform.**

   **Buffer X2:**

   20 mM Tris·Cl (pH 8.0)
   20 mM EDTA
   200 mM NaCl
   80 mM DTT
   4% SDS
   250 µg/ml Proteinase K

   Just before use, add the appropriate volume of Proteinase K stock solution (20 mg/ml) supplied with the QIAamp DNA Mini Kit.

   DTT oxidizes quickly in aqueous solutions and should also be added just before use. Store the DTT stock solution (1 M) at –20°C.

2. **Add 200 µl Buffer AL and 200 µl ethanol to the sample. Mix by vortexing.**

3. **Continue with step 5 to step 8 of the Tissue Protocol in the *QIAamp DNA Mini Kit and QIAamp DNA Blood Mini Kit Handbook.***

4. **Elute the DNA in 50–100 µl Buffer AE or distilled water.**

   **Note:** Elution in 50 µl will yield more concentrated DNA, whereas elution in 100 µl will recover a greater amount of DNA. If the expected amount of DNA is not known, it is preferable to elute in several aliquots of 50 µl. These can then be combined if necessary.

   Elution of the DNA in Buffer AE is recommended if the DNA is to be stored, since DNA stored in water is subject to acid hydrolysis.

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