

## Storage of Blood Samples Collected into PAXgene™ Blood DNA Tubes at –80°C

**Experimental design:** Human whole blood samples from 15 donors were drawn into PAXgene Blood DNA Tubes (10 samples per donor, 150 samples in total) and stored at –80°C for up to 24 months, as part of an ongoing study.

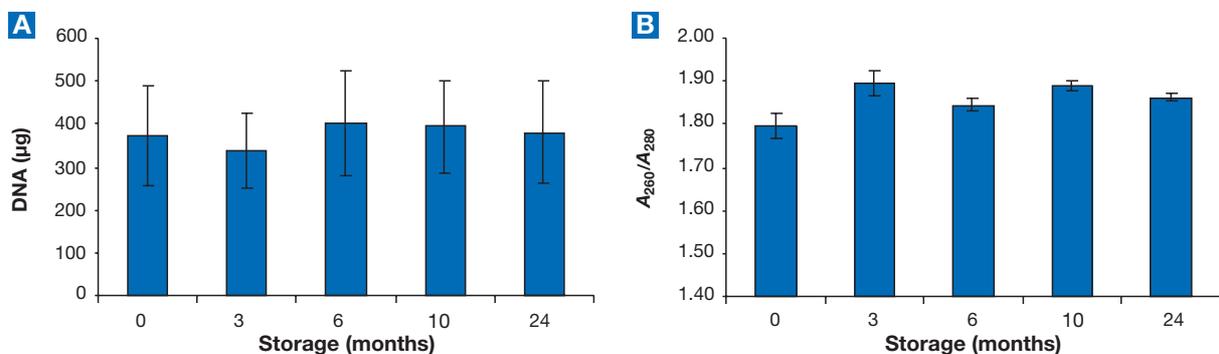
Duplicate samples from each donor were processed on day 0. Remaining samples were immediately frozen at –80°C (samples from 15 donors) horizontally in a plastic bag. Duplicate samples from each donor were processed after storage for 3 months and 6 months, and one sample from each donor was processed after storage for 10 months and 24 months. Samples were processed using the PAXgene Blood DNA Kit according to the standard protocol. The DNA was dissolved in 1 ml Buffer BG4 (resuspension buffer).

**Analytical methods:** In total, 120 blood samples (30 at day 0, 30 each after 3 and 6 months, and 15 each after 10 and 24 months) were analyzed. Amount and purity of DNA were analyzed by measuring the absorbance at 260 and 280 nm ( $A_{260}/A_{280}$ ). In addition the purified DNA was analyzed by agarose gel electrophoresis and by PCR amplification of a 1.1 kb fragment of the human single-copy gene *Hugl* (human homolog of giant larvae).

**Results:** The average DNA amount remained unchanged within statistical limits after 2 years of storage with 374 µg on day 0, 337 µg after 3 months, 403 µg after 6 months, 393 µg after 10 months, and 381 µg after 24 months (Figure 1A). DNA purity remained high in all samples tested, with the  $A_{260}/A_{280}$  ratio consistently between 1.7 and 1.9 (Figure 1B).

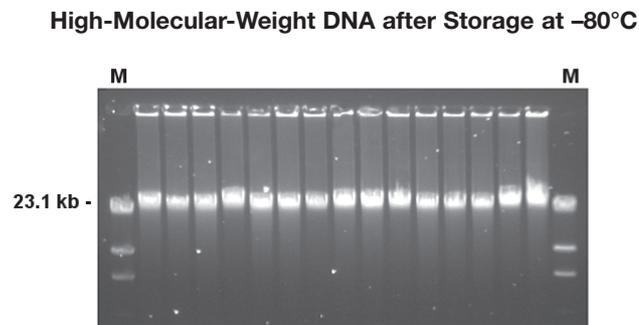
Agarose gel analysis showed that after 24 months of storage at –80°C, DNA samples still ran quantitatively above a 23 kb marker band (Figure 2). In addition, a 1.1 kb fragment of the human single-copy gene *Hugl* was successfully amplified from all DNA samples (Figure 3).

### Average Amount and Purity of DNA after Storage in PAXgene Blood DNA Tubes at –80°C for up to 24 Months



**Figure 1.** **A** Average amount and **B** average purity of DNA purified from blood samples from 15 donors before and after storage for 24 months at –80°C. **Note:** High interdonor variation of number of nucleated cells leads to high standard deviation of DNA yield.

**Conclusion:** High-quality, highly concentrated genomic DNA can be isolated from blood samples after 24 months of storage at  $-80^{\circ}\text{C}$  using the PAXgene Blood DNA System.



**Figure 2.** Agarose gel analysis (0.7% agarose gel, 1x TAE buffer, 80 V, 5 hours) of 400 ng DNA purified from blood samples from 15 donors after storage in PAXgene Blood DNA Tubes for 24 months at  $-80^{\circ}\text{C}$ . **M:** markers.



**Figure 3.** Amplification of a 1.1 kb fragment of the single-copy gene Hugl. DNA was purified from blood samples from 15 donors after storage in PAXgene Blood DNA Tubes for 24 months at  $-80^{\circ}\text{C}$ . **M:** markers; **W:** water control.

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The PAXgene Blood DNA System is for research use only and not for use in diagnostic procedures.

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.

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