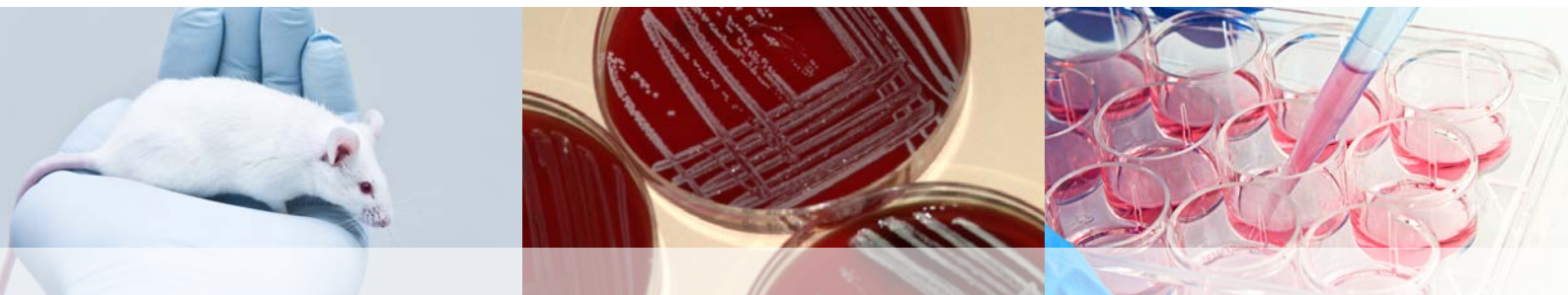


NucleoSpin® 8 RNA

RNA isolation from PAXgene® Blood RNA Tubes using NucleoSpin® 8 RNA



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Abstract

In this application note we show the high throughput extraction of RNA from blood samples stored in PAXgene® Blood RNA Tubes. RNA was extracted from human blood samples after storage using NucleoSpin® 8 / 96 RNA kit on NucleoVac Vacuum Manifold. RNA yield and quality was analyzed with the Agilent 2100 Bioanalyzer.

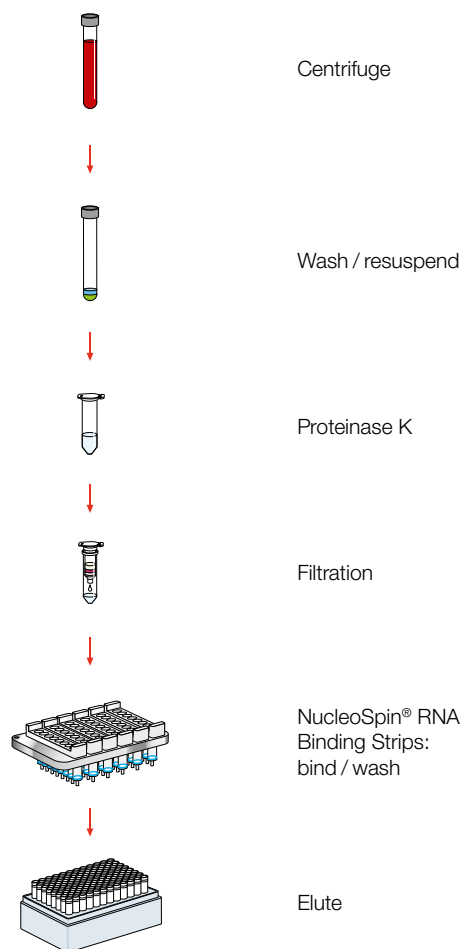
Introduction

For RNA isolation from human blood samples, RNA quality is of highest interest. Preserving RNA quality in blood samples for transcriptome analysis, the PAXgene® Blood RNA Tube is often used. Blood samples can be stored at room temperature for several days without losing RNA quality. This application note is describing the use of the MACHEREY-NAGEL kit NucleoSpin® 8 RNA for RNA isolation from PAXgene® Blood RNA Tubes. A detailed protocol of the procedure can be requested at tech-bio@mn-net.com.

Material and methods

To test the compatibility of PAXgene® Blood RNA Tubes with NucleoSpin® 8 / 96 RNA kit, six human blood samples were used. The processing of PAXgene® Blood RNA Tubes (2.5 mL blood) was performed according to the manufacturer's protocol.

After storage at 4 °C, the samples were equilibrated to room temperature for RNA isolation. First the PAXgene® Tube was centrifuged at 3000–5000 x *g* for 10 min, after removing the supernatant the pellet was resuspended in 5 mL RNase-free water for washing. After washing the pellet was resuspended in 300 µL RNase-free water and Lysis Buffer RA1 (+ TCEP or β-mercaptoethanol) and Proteinase K were added. The lysate was cleared with the NucleoSpin® Filter, afterwards 300 µL ethanol was added and the sample was loaded onto the NucleoSpin® 8 RNA binding strip. After binding step the standard NucleoSpin® 8 RNA protocol was performed.



Results

The RNA yield from 2.5 mL human blood samples was about 4 µg. The RNA integrity tested with the Agilent Bioanalyzer showed RIN values of 8 indicating high RNA integrity without significant degradation.

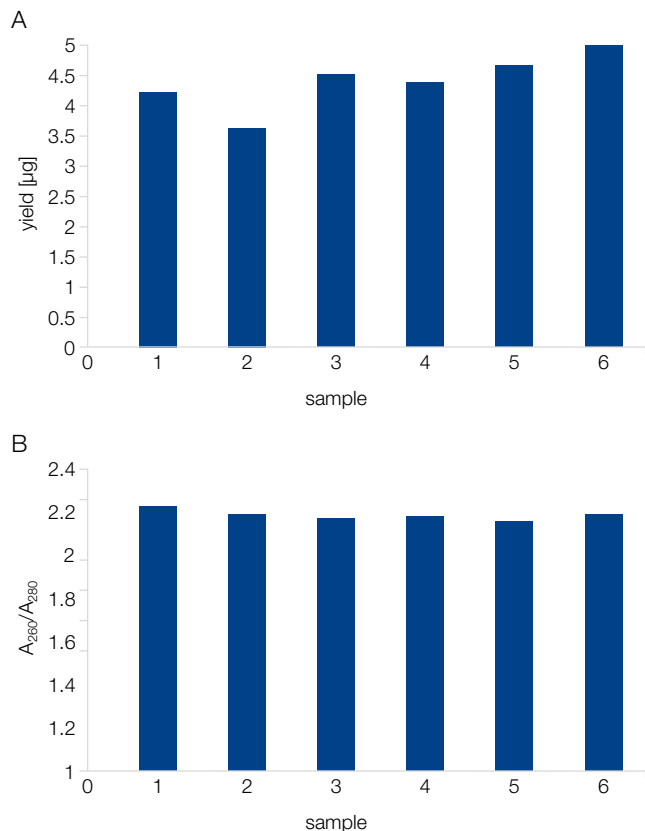


Figure 1: RNA isolation from 2.5 mL human blood samples, stabilized in PAXgene® Blood RNA Tubes. The graphs show RNA yield (A) and A_{260}/A_{280} ratio (B) of three analyzed samples in duplicate.

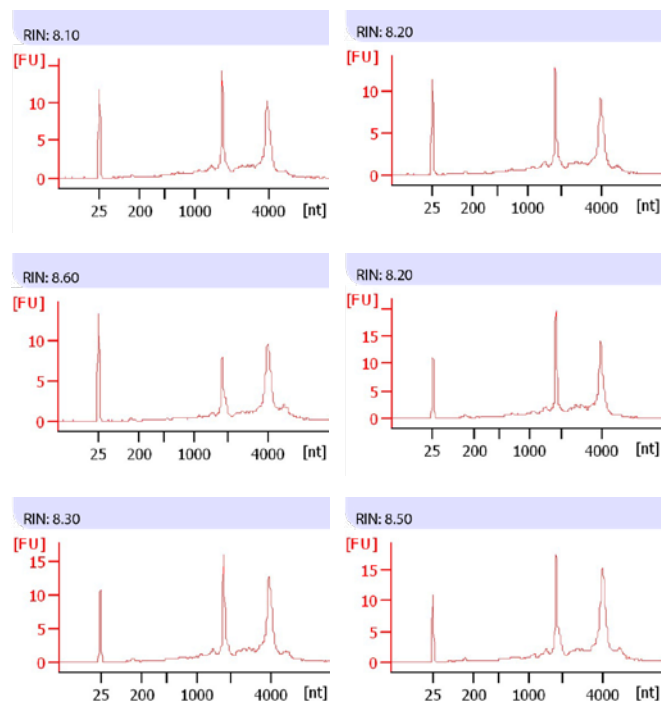


Figure 2: Electropherograms obtained from the Agilent Bioanalyzer 2100 using RNA Nano 6000 reagents. RNA was isolated from 2.5 mL blood. RNA integrity numbers (RIN) are above 8 showing a high RNA integrity.

Conclusion

PAXgene® Blood RNA Tubes can be processed for RNA isolation with MACHERY-NAGEL NucleoSpin® 8 RNA kit. Purified RNA is of high integrity and quality for optimal performance in typical downstream applications.

Besides PAXgene® Blood RNA tubes, blood samples are often stored in standard blood collection tubes (with EDTA, heparin, or citrate). For the RNA purification from EDTA, citrate, or heparin blood samples the use of NucleoSpin® 8/96 RNA Blood kit is recommended.

For further details please contact tech-bio@mn-net.com or visit www.mn-net.com/Bioanalysis.

Ordering information

Product	Preps	REF
NucleoSpin® 8 RNA	12 x 8	740698
	60 x 8	740698.5
NucleoSpin® 96 RNA	2 x 96	740709.2
	4 x 96	740709.4
NucleoSpin® Filters	50	740606

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