Easy purification of high-performance DNA from paraffin-embedded tissue sections using the MagAttract[®] DNA Mini M48 Kit

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This study describes the efficient purification of DNA from paraffin-embedded sections of human bone marrow, cervix, lymphatic, and skin samples. DNA was purified from paraffin-embedded tissue sections using the MagAttract® DNA Mini M48 Kit and the BioRobot® M48 workstation. The performance of DNA purified from paraffin-embedded human tissue was compared by PCR to performance of control DNA.

Purification of genomic DNA from paraffin-embedded sections can take up to 5 days, involves the use of hazardous organic solvents, and often yields nicked or otherwise degraded DNA. A standardized, automated method to purify DNA from paraffin-embedded tissues for PCR analyses would help conserve precious samples, reduce contact with hazardous chemicals, and accelerate genotyping studies.

Materials and methods

A range of paraffin-embedded tissue sections (5–10 x 10 µm-thick sections) were used per sample, dependent on the size and cell-content of the sections. Samples were placed directly in 190 µl Buffer G2 together with 10 µl Proteinase K, and incubated at 56°C overnight with mixing. Following incubation, samples were processed using the BioRobot M48 workstation and purified DNA was eluted in 100 µl water and quantified by absorbance (A_{260}), corrected for background (A_{320}).

The quality of the purified DNA was tested by PCR. Three fragments (266, 167, and 85 bp) were amplified from discrete regions of the human serum albumin gene using 25 ng of purified DNA. DNA quality was assessed using the Agilent 2100 bioanalyzer. Bioanalyzer scans of test PCRs were compared with a control PCR using control DNA.

Results

The yield (Table 1) and quality of the purified DNA was variable, as expected from formalin fixed tissues submitted for routine pathology testing. Analysis of PCR (Figure 1) showed that the purified DNA performed well in sensitive multiplex PCR.

Table 1. Functional DNA Yields from a Rangeof Paraffin-Embedded Human Tissues

Yield (µg)	Concentration (ng/µl)
4.4 ± 1.4	44.4 ± 14.2
6.8 ± 3.4	68.3 ± 33.9
3.2 ± 2.4	32.2 ± 23.8
5.9 ± 3.2	59.0 ± 31.7
	4.4 ± 1.4 6.8 ± 3.4 3.2 ± 2.4



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Specific PCR using DNA from Paraffin-Embedded Tissues

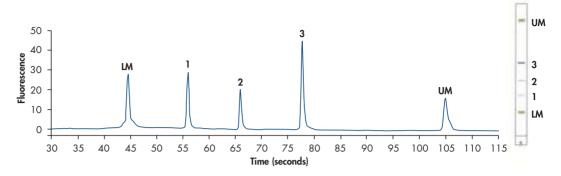


Figure 1 Bioanalyzer analysis of a PCR of 3 regions (266, 167, and 85 bp) of the human serum albumin gene. This figure shows a result generated using high-quality purified DNA. **LM**: lower marker; **UM**: upper marker; **1**: 86 bp fragment; **2**: 167 bp fragment; **3**: 266 bp fragment.

Conclusions

These results clearly demonstrate that using the MagAttract DNA Mini M48 Kit in combination with the BioRobot M48 workstation provides functional yields of high-performance DNA from paraffin-embedded sections.

Ordering Information

Product	Contents	Cat. no.
MagAttract DNA Mini M48 Kit (192)	For 192 DNA preps: MagAttract Suspension B, Buffers, Proteinase K	953336
BioRobot M48	Robotic workstation for automated purification of nucleic acids using MagAttract M48 Kits, installation, 1-year warranty on parts and labor	9000708

Trademarks: QIAGEN[®], BioRobot[®], MagAttract[®] (QIAGEN Group). QIAGEN Robotic Systems are not available in all countries; please inquire. The BioRobot M48 and MagAttract Kits are intended as general purpose devices. No claim or representation is intended for their use in identifying any specific organism or for a specific clinical use. It is the user's responsibility to validate the performance of the BioRobot M48 and MagAttract Kits for any particular use, since performance characteristics have not been validated for any specific organism. The BioRobot M48 and MagAttract Kits may be used in clinical diagnostic laboratory systems their after the laboratory has validated their complete system as required by CLIA '88 regulations in the U.S. or equivalents in other countries. 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. 01/2004 © 2004 QIAGEN, all rights reserved.

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