# Automated Isolation of DNA from Tissue Samples in 35–50 Minutes

## Fast and Easy Purification Combining the MagNA Lyser and the MagNA Pure Compact Systems

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## Introduction

The efficient isolation of DNA from various tissues is crucial in *e.g.*, mutation analysis or gene quantification. Roche Applied Science has developed new kits and protocols for automated isolation of genomic DNA using the MagNA Lyser System and the MagNA Pure Compact System.

The MagNA Lyser (Figure 1a) is a small benchtop instrument that allows the efficient homogenization of up to 16 tissue samples within only a few seconds. It is based on a bead-beating technology, and uses disposable 2-ml tubes with ceramic beads that are vigorously shaken, using up to 7,000 rpm to disrupt the tissue.

The MagNA Pure Compact System consists of a fully automated benchtop instrument (Figure 1b) and ready-to-use nucleic acid isolation kits with prefilled cartridges, allowing automated isolation of nucleic acids from 1–8 samples within 25–40 minutes. The system has an integrated touchscreen computer and a bar-code reader. It comprises full sample tracking and liquid, clot, and tip detection.

The combination of the two systems results in a fast and efficient process, allowing the isolation of DNA from various tissue samples in less than one hour with minimum hands-on time.

## Materials and Methods

### Homogenization of tissue

For one isolation, 1–10 mg of mouse tissue (liver, kidney, spleen, lung, ear, muscle, heart, brain) were homogenized using the MagNA Lyser Instrument and the MagNA Lyser Green Beads tubes together with 100–400 µl MagNA Pure DNA Tissue Lysis Buffer. In general, homogenization was done at 6000 rpm for 30–40 seconds, in case of pertinacious tissues (ear, muscle) for 2 x 30 seconds. For comparison, an overnight digestion with proteinase K was performed in parallel.

### Optional RNase digestion step

RNase A solution (*i.e.*, 10 µl of a 40 mg/ml solution) was added to the lysate and incubated for 10–15 minutes at 65°C to eliminate RNA and to prevent its co-isolation from tissues containing large amounts of RNA (*e.g.*, liver).

### Isolation on the MagNA Pure Compact System

Samples were transferred to the MagNA Pure Compact System and automated DNA isolation was started using the MagNA Pure Compact Nucleic Acid Isolation Kit Large Volume together with the DNA-Cells Isolation Protocol. This optimized protocol automatically performs all DNA isolation steps.

### Analysis

DNA isolated by this method was analyzed by agarose gel electrophoresis, OD measurements and PCR using the LightCycler® Instrument.

## Results and Discussion

Table 1 shows the DNA yield (as determined by $OD_{260\,nm}$ measurement) and purity (ratio $OD_{260\,nm/280\,nm}$) obtained from various tissue types. In most cases, the rapid MagNA Lyser treatment gave similar or higher yields than

**Figure 1:** (*a*) MagNA Lyser Instrument (*b*) MagNA Pure Compact Instrument

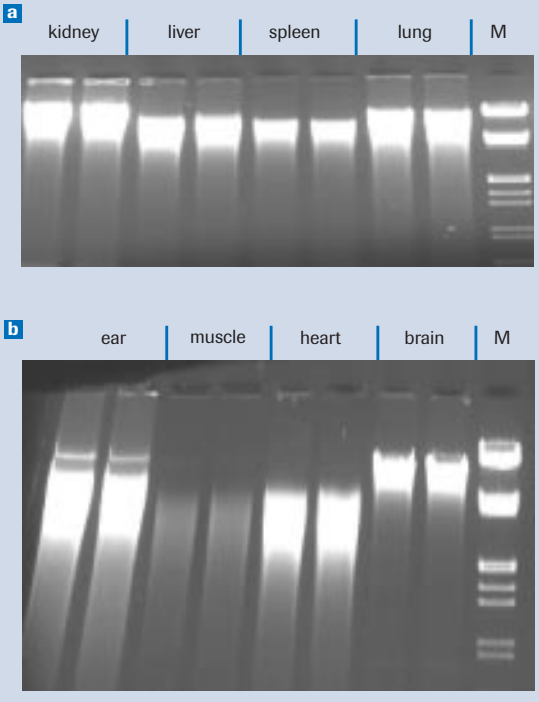
**Table 1: DNA yield and purity from various tissues.** Homogenization was done with the MagNA Lyser and for comparison by overnight digestion with proteinase K.

| Tissue type | Amount | DNA yield [µg] (MagNA Lyser) | DNA yield [µg] (proteinase K) | Ratio $OD_{260nm}/OD_{280nm}$ |
|---|---|---|---|---|
| Liver | 10 mg | 23.4 | 20.5 | 1.9 |
| Spleen | 10 mg | 47.2 | 15.1 | 1.9 |
| Kidney | 10 mg | 23.3 | 28.1 | 1.9 |
| Lung | 10 mg | 24.2 | 17.5 | 1.9 |
| Ear | 10 mg | 5.4 | 18.7 | 1.9 |
| Muscle | 10 mg | 4.7 | 4.4 | 1.9 |
| Heart | 10 mg | 9.7 | 16.6 | 1.9 |
| Brain | 10 mg | 7.5 | 13.4 | 1.9 |

overnight digestion with proteinase K. The latter, however, can be advantageous for difficult tissues such as heart, ear/skin, or brain tissue. Yields were generally higher than those obtained with other isolation methods (data not shown).

Figures 2 a and b show the results of the agarose gel analysis. The DNA was of good integrity. Some smear due to the homogenization is normally observed. The average size was between 2,000 kb and 20,000 kb, depending on the tissue type and the duration of the MagNA Lyser treatment.

Figure 3 shows a PCR analysis using the LightCycler® Instrument. The isolated DNA from various types of mouse tissue was amplified using cyclophilin A as target gene.




**Figures 2: (*a, b*) Agarose gel analysis of DNA isolated from various tissues using MagNA Lyser and MagNA Pure Compact.** All isolations were done in duplicates. Eluates were then analyzed on a 1% agarose gel together with DNA Molecular Weight Marker III. The DNA of some of the samples (*e.g.*, muscle, heart) is partially fragmented due to a more extensive mechanical homogenization.

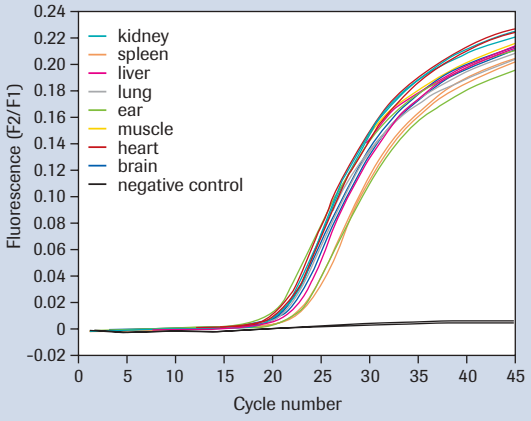



**Figure 3: LightCycler® System analysis of DNA from mouse tissue.** Cyclophilin A was chosen as a target gene for quantitative real-time PCR using the LightCycler® Instrument.

Five microliters of eluate per PCR were used. The curves show that the isolated DNA was amplified within the expected crossing-point range and no signs of inhibition were found.

## Conclusion

The combination consisting of the MagNA Lyser System and the MagNA Pure Compact System proved to be a very efficient tool for automated isolation of DNA from various tissue types. Rapid tissue homogenization together with rapid automated purification allows the isolation of pure DNA from tissues within 35–50 minutes.

The yield and purity of the nucleic acids was high as shown by agarose gel electrophoresis, by OD analysis and by amplification in the LightCycler® Instrument. This workflow significantly simplifies the molecular analysis of various kinds of research tissue samples. ■

Order INFO

| Product | Pack Size | Cat. No. |
|---|---|---|
| **MagNA Lyser Instrument** | 1 instrument | 03 358 968 001 |
| **MagNA Lyser Green Beads** | 100 tubes | 03 358 941 001 |
| **MagNA Pure DNA Tissue Lysis Buffer** | 100 ml | inquire |
| **RNase A** | 25 mg<br>100 mg | 10 109 142 001<br>10 109 169 001 |
| **MagNA Pure Compact Instrument** | 1 instrument | 03 731 146 001 |
| **MagNA Pure Compact Nucleic Acid Isolation Kit I, Large Volume** | 32 isolations | 03 730 972 001 |
| **DNA Molecular Weight Marker III** | 50 µg | 10 528 552 001 |