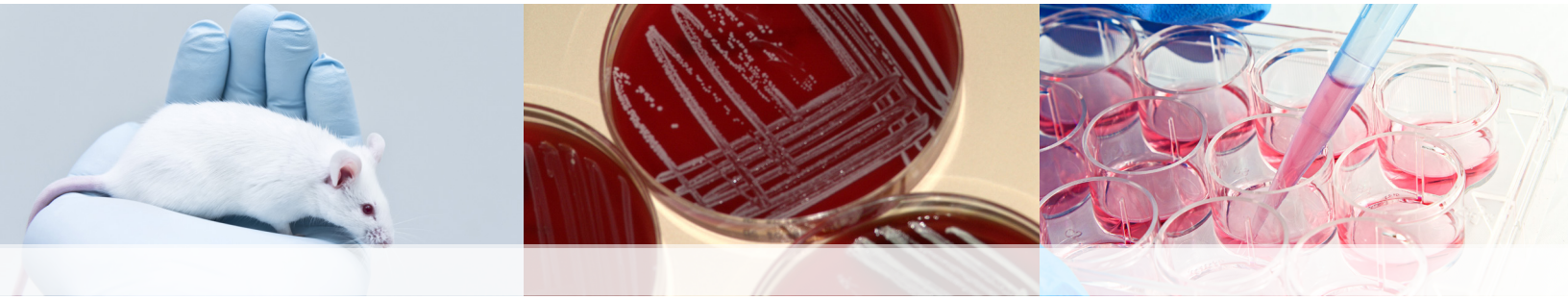
MACHEREY-NAGEL

NucleoSpin® 96 DNA RapidLyse

Automated purification of genomic DNA from tissue or cells using the Hamilton® [MPE]^2 positive pressure module



Introduction

The isolation of genomic DNA from tissues like mouse tails, mammalian organs, or even eukaryotic cells can be a time consuming task. However, efficient lysis and DNA release is essential for subsequent downstream molecular applications, utilized by many research laboratories. MACHEREY-NAGEL designed the silica membrane based NucleoSpin® 96 DNA RapidLyse kit with a unique buffer chemistry to enable a shortened cost efficient purification workflow.

Sample lysis can be performed in 15–60 min depending on sample material and high quality DNA can be extracted and directly used as a template for PCR, NGS, blotting, or various other enzymatic reactions. This silica membrane based kit can be successfully used with either centrifugation or standard vacuum processing in manual or automated manner.

MACHEREY-NAGEL is continuously expanding on its collaborations with automation partners in order to offer more support to high throughput customers. We now present the first implementation of the NucleoSpin® 96 DNA RapidLyse kit on a positive pressure unit using the [MPE]^2 positive pressure module from Hamilton®. The [MPE]^2 module maintains equal pressure across the NucleoSpin® 96 DNA RapidLyse Binding Plate eliminating path of least resistance. Our optimized protocol allows the processing of 96 samples within approximately 60–90 minutes, depending on platform setup.

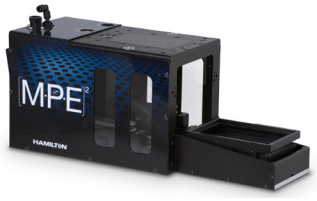
Product at a glance

NucleoSpin® 96 DNA RapidLyse	
Technology	Silica membrane technology
Sample material	≤ 30 mg tissue, < 10^6 cells
Preparation time	Approx. 60–90 min depending on platform setup (excl. lysis of 15–60 min)
Typical yield	Up to 4 µg DNA/mg tissue or 10^6 cells
Elution volume	100 µL
Theoretical binding capacity	40 µg

[MPE]^2	
Technology	Monitored Multi-flow, Positive Pressure Evaporative Extraction
Sample volume	Optional reagent fill module with up to 15 reagent bottles
Capacity	24 / 48 / 96 samples
Size / weight	44.5 x 15.9 x 18.1 cm / 6.9 kg

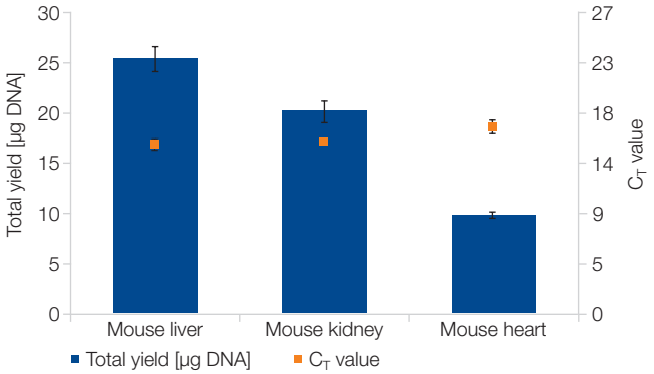
Material and methods

Samples from mouse or pig organs (20–30 mg tissue) and 1 x 10^5 / 10^6 cells were lysed in maximal one hour agitated incubation at 56 °C. The highly efficient DNA release is enabled by a thoroughly designed lysing setup with optimized parameters that comprise the special Lysis Buffer RLY in combination with Liquid Proteinase K. Incubation overnight or for several hours is not necessary. Following lysis and the addition of Binding Buffer RLB, all subsequent steps are performed on the [MPE]^2 positive pressure module.



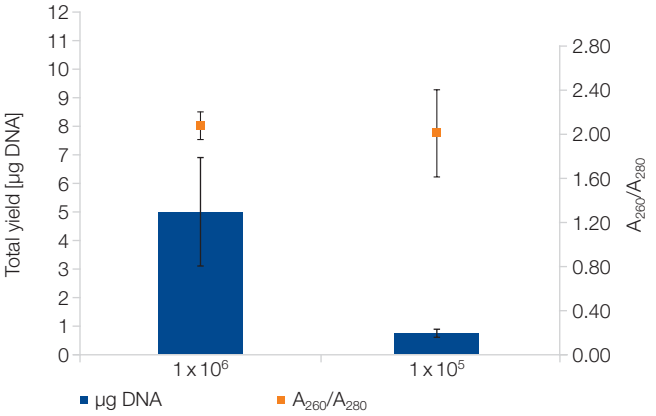
Nucleic acids are reversibly bound to the silica membrane during the binding step. Contaminants, such as salts or lipids, are then removed from the silica membrane by three washing steps using Washing Buffer RLW, while nucleic acids stick to the silica membrane. Highly pure genomic DNA is finally eluted under low ionic strength conditions in a slightly alkaline Elution Buffer RLE.

Application data



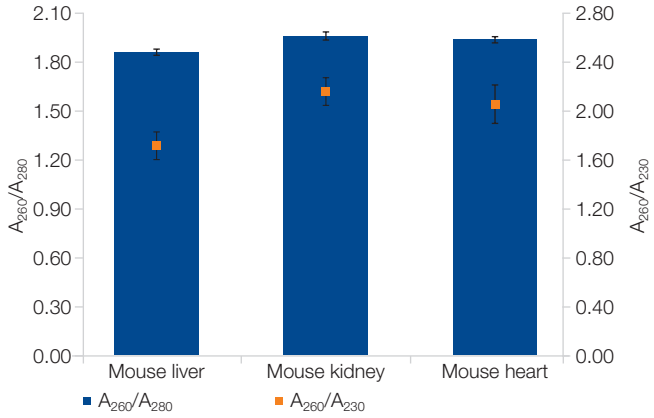
Automated isolation of genomic DNA from mouse organs

DNA was isolated from various mouse tissue samples (n= 8, 30 mg each) using the NucleoSpin® 96 DNA RapidLyse kit on a [MPE]² positive pressure module. The total yield was determined by UV spectrometry (dark blue bars). A subsequent qPCR analysis (orange squares) was performed with a Taqman® Probe for a GADPH amplicon using the SensiFast™ Probe Lo-ROX kit from Bioline on an Applied Biosystems® 7500 Real-Time PCR System.



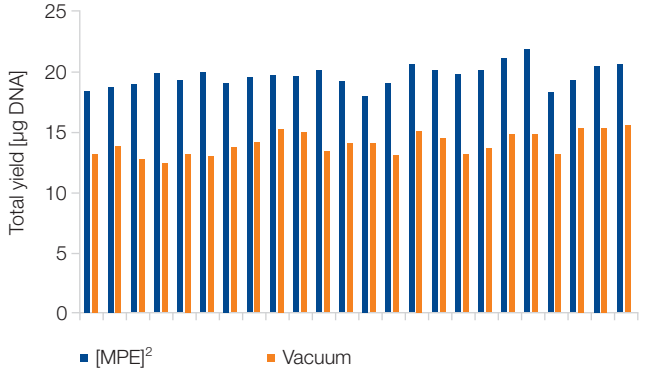
Yield and purity of isolated genomic DNA from HeLa cells

DNA was isolated from different amounts of HeLa cells (n = 8,) using the NucleoSpin® 96 DNA RapidLyse kit on a [MPE]² module. The total yield was determined by UV spectrometry (dark blue bars). Purity measurement (orange squares) was performed by UV spectrometry, resulting in an average A_{260}/A_{280} value for 1x10^6 HeLa cells = 2.02 ± 0.28 and for 1x10^5 HeLa cells = 1.94 ± 0.38.



Purity of isolated genomic DNA from mouse organs

DNA was isolated from various mouse tissue samples (n = 8, 30 mg each) using the NucleoSpin® 96 DNA RapidLyse kit on a [MPE]² positive pressure module. The purity was determined by UV spectrometry, resulting in an average A_{260}/A_{280} value for liver tissue of 1.89 ± 0.02; for kidney tissue of 1.96 ± 0.02; for heart tissue of 1.96 ± 0.01 (dark blue bars). The average A_{260}/A_{230} value for liver tissue = 1.72 ± 0.12 for kidney tissue = 2.17 ± 0.12; for heart tissue = 2.04 ± 0.22 (orange squares).



Comparison of positive pressure and vacuum processing

DNA was isolated from pig kidney samples (n = 24; 20 mg each) using the NucleoSpin® 96 DNA RapidLyse kit on the [MPE]² positive pressure module (blue bars) or on the NucleoVac 96 vacuum manifold (orange bars). Moreover, the purity was determined by UV spectrometry, resulting in an average A_{260}/A_{280} value for positive pressure processing of 1.93 ± 0.03 and for vacuum processing of 1.95 ± 0.02. The average A_{260}/A_{230} value for positive pressure processing = 2.18 ± 0.74 and for vacuum processing = 1.93 ± 0.29.

Speed up and automate your gDNA extraction from e.g., tissue samples and cells

MACHEREY-NAGEL and Hamilton® deliver a sophisticated solution for high throughput DNA extraction. The NucleoSpin® 96 DNA RapidLyse procedure can be easily adapted to the [MPE]² positive pressure module and speed up your DNA extraction workflow.

- Reliable performance and excellent yields using NucleoSpin® 96 DNA RapidLyse on the [MPE]² positive pressure module
- Compact automated processing of 96 samples in 60–90 minutes (excluding lysis)

Ordering information

Product	Specifications	Preps	REF
NucleoSpin® 96 DNA RapidLyse	Genomic DNA purification kit for a variety of sample materials in 96-well format	1 x 96 / 4 x 96	740110.1 / .4
MN Wash Plate	Plate to minimize the risk of cross-contamination	4 / 24	740479 / .24
[MPE]²	Monitored multi-flow, positive pressure evaporative extraction module with 96 air manifold and evaporator		96160–04*

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