MACHEREY-NAGEL NucleoSpin[®] 96 Tissue

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



Introduction

The efficient isolation of genomic DNA from a variety of tissue samples and/or cell types is essential for subsequent downstream molecular applications utilized by many research laboratories. MACHEREY-NAGEL designed the NucleoSpin[®] 96 Tissue kit for the rapid and automated parallel purification of genomic DNA from tissue samples or cells in a 96-well format. 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.

MN is continuously expanding the collaborations with automation partners in order to offer more support to high throughput customers. We now present the first implementation of the NucleoSpin[®] 96 Tissue 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[®] Tissue Binding Plate eliminating path of least resistance. Our optimized protocol allows the processing of 96 samples within approximately 60 to 90 minutes, depending on platform setup.

Product at a glance

NucleoSpin [®] 96 Tissue				
Technology	Silica membrane technology			
Sample material	\leq 20 mg tissue, $< 10^6$ cells			
Preparation time	Approx. 60–90 min depending on platform setup (excl. lysis)			
Typical yield	15–25 µg (20 mg tissue)			
Elution volume	100–200 µL			
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 up to 20 mg tissue, 1 x 10^6 cells or bacterial pellets are lysed with Lysis Buffer T1 and Proteinase K for 1–16 h at 56°C. Lysis incubation time depends on sample type (see the NucleoSpin[®] 96 Tissue user manual for more detailed information). Following lysis and the addition of binding buffer, all subsequent steps are performed on the [MPE]² positive pressure module.



After sample lysis the nucleic acids are reversibly bound to the silica membrane of the NucleoSpin[®] 96 Tissue Binding Plate. An optimized protocol and plate stack assembly enables the purification of highly pure genomic DNA using the MN Wash plate. The MN Wash Plate is a microtiter plate open on both sides thus allowing free flow-through generating 96 separate channels

During the washing steps of the developed protocol the MN Wash Plate is placed underneath the [MPE]² Adapter frame. The NucleoSpin[®] 96 Tissue Binding Plate is placed on top of the [MPE]² Adapter frame. This plate stacking prevents the bottom of the NucleoSpin[®] 96 Tissue Binding Plate to be contaminated and the flow through is drained away from the plate directly into the waste reservoir. Contaminants, such as salts or lipids, are then removed from the silica membrane by three washing steps, while nucleic acids are reversible bound to the silica membrane. DNA is finally eluted under low ionic strength conditions in a slightly alkaline Elution Buffer (BE).



Application data



Automated isolation of genomic DNA from mouse tail samples

DNA was isolated from mouse tail samples (n= 16, 20 mg each) using the NucleoSpin[®] 96 Tissue kit on a [MPE]². 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 on an Applied Biosystems[®] 7500 Real-Time PCR System. The comparable results demonstrate consistently high DNA yield for all tested samples.



Purity of isolated genomic DNA from mouse tail samples

DNA was isolated from mouse tail samples (n = 16, 20 mg each) using the NucleoSpin[®] 96 Tissue kit on a [MPE]² positive pressure module. The purity was determined by UV spectrometry, resulting in an average A₂₆₀/A₂₈₀ value of 1.95 ± 0.02 (dark blue bars) and an average A₂₆₀/A₂₃₀ value of 2.31 ± 0.1 (orange squares).



Integrity of isolated DNA

The integrity of the isolated nucleic acids from mouse tail samples was analyzed by gel electrophoresis (7.5 μ L per eluate; 0.7 % TAE gel; M: Lamda DNA/Hind III – Thermo Scientific)



Comparison of positive pressure and vacuum processing

DNA was isolated from deer liver samples (n = 16; 20 mg each) using the NucleoSpin[®] 96 Tissue kit on the [MPE]² or on the NucleoVac 96 vacuum manifold. The total yield was determined by UV spectrometry resulting into an average total yield of 10.17 \pm 0.96 μg (dark blue bars) for positive pressure processing or 9.54 \pm 1.48 μg (light blue bars) for vacuum processing.

Automate your DNA extraction from tissue samples and cells

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

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

Product	Specifications	Preps	REF
NucleoSpin [®] 96 Tissue	Kit based on silica membrane technology for the isolation of genomic DNA from tissue samples in 96-well format	2 x 96 4 x 96 24 x 96	740741.2 740741.4 740741.24
MN Wash plate	Plate to minimize the risk of cross-contamination	4 24	740479 740479.24
Elution plate U-bottom	96-well microplate with 300 µL u-bottom, including self-adhering foil	24	740486.24
[MPE] ²	Monitored multi-flow, positive pressure evaporative extraction module with 96 air manifold and evaporator		96160-04*

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