MACHEREY-NAGEL

NucleoSpin® 96 DNA RapidLyse

Automated purification of DNA from various tissues using the platform ep*Motion*® 5075vt

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 of 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 on the automation platform in 15–60 min depending on sample material. High quality DNA can be extracted on the ep*Motion*® 5075vt 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.

This application note describes the automated process on the automated liquid handling workstation ep*Motion*® 5075vt using the NucleoSpin® 96 DNA RapidLyse kit from MACHEREY-NAGEL. The novel optimized protocol allows the processing of variable sample numbers in multiples of 8 (8–96). The processing of 96 samples takes approximately 104 minutes excluding sample lysis.

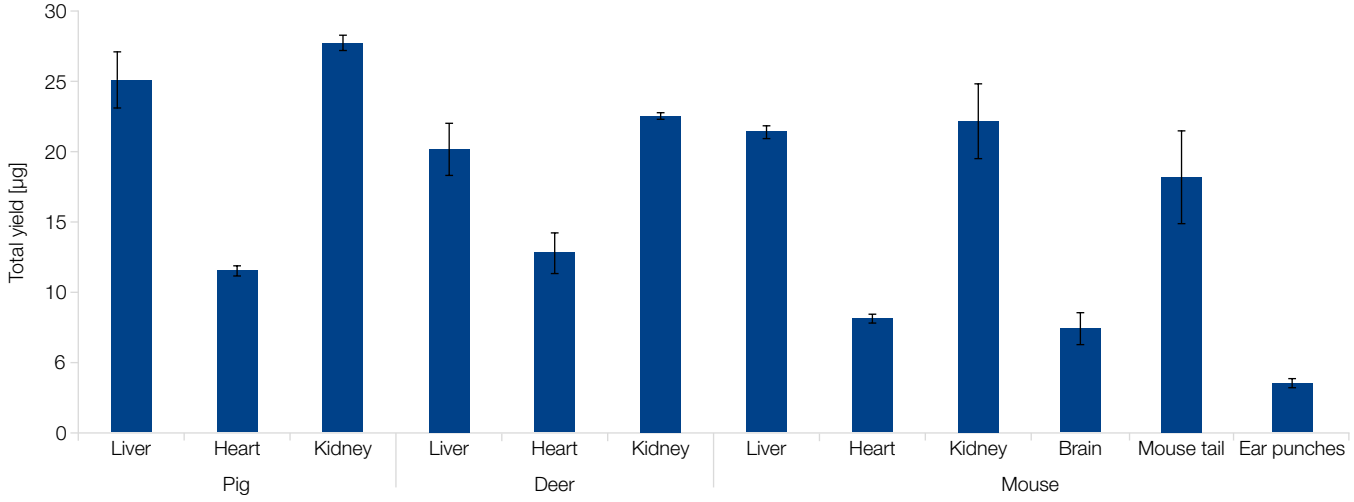
Product at a glance

NucleoSpin® 96 DNA RapidLyse	
Technology	Silica membrane technology
Sample material	≤ 30 mg tissue, < 10^6 cells
Preparation time	Approx. 104 min for 96 samples (excluding lysis)
Format	Variable sample number in multiples of 8 (8–96)
Typical yield	Up to 4 µg DNA/mg tissue or 10^6 cells
Elution volume	100 µL
Theoretical binding capacity	40 µg

Material and methods

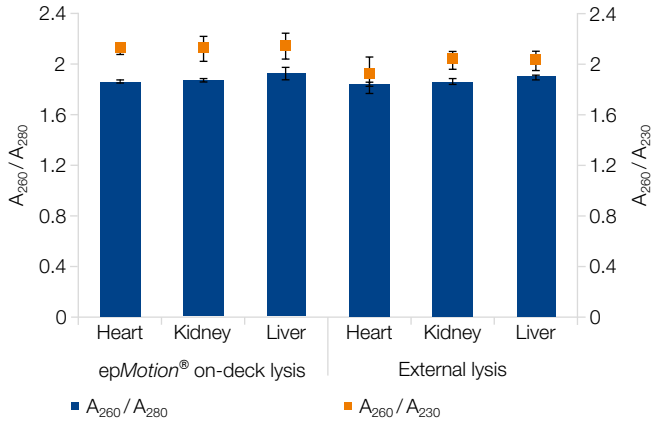
The optimized protocol is programmed to process up to 96 samples in parallel (variable sample number in multiples of 8) and developed for the ep*Motion*® 5075vt platform. Fresh or frozen blood samples, treated with EDTA as anticoagulant, and may be derived from humans or animals (e.g., pig or cow). Samples from e.g., mouse, deer or pig organs (20–30 mg tissue) and ear punches (~12 mg) 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. 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



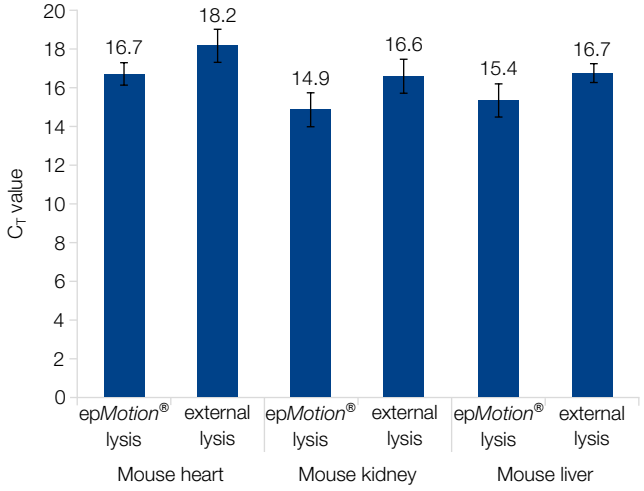
Automated isolation of genomic DNA from pig, deer and mouse organs

DNA was isolated from various animal tissue samples (n = 8, 30 mg each tissue organ; ~12 mg each mouse ear punch) using the NucleoSpin® 96 DNA RapidLyse kit on a ep*Motion*® 5075vt platform. The total yield was determined by UV spectrometry (dark blue bars).



DNA purity from mouse organs depending on external or on-deck lysis

DNA was isolated from various mouse tissue samples (n = 8, 30 mg each) using the NucleoSpin® 96 DNA RapidLyse kit on ep*Motion*® 5075vt platform. The purification workflow was processed with either performing ep*Motion*® 5075vt on-deck lysis (ODL) or an external manual lysis step (EML). The purity was determined by UV spectrometry.



DNA yield from mouse organs depending on manual or on-deck lysis

DNA was isolated from different mouse tissue samples (n = 8, 30 mg each) using the NucleoSpin® 96 DNA RapidLyse kit on ep*Motion*® 5075vt platform. The purification workflow was processed with either performing ep*Motion*® 5075vt on-deck lysis (ODL) or an external manual lysis step (EML). A subsequent qPCR analysis was performed with a Taqman® Probe for a GAPDH amplicon using the SensiFast™ Probe Lo-ROX kit from Bioline on an Applied Biosystems® 7500 Real-Time PCR System.

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

MACHEREY-NAGEL and Eppendorf® deliver a fully automated solution for your high throughput DNA extraction from various tissues

- Fully automated DNA extraction including on deck lysis and subsequent purification using NucleoSpin® 96 DNA RapidLyse
- Flexible sample numbers (multiple of 8) and fast processing of 96 samples within 104 minutes (excluding lysis)

Ordering information

Product	Specifications	Preps	REF
NucleoSpin® 96 DNA RapidLyse	Kit based on silica membrane technology for fast isolation of genomic DNA from a variety of sample materials in 96-well format	1 x 96 / 4 x 96	740110.1 / .4
ep*Motion*® 5075vt	Basic device incl. vacuum system, gripper, vac frame 2, vac frame holder, Eppendorf ThermoMixer®, epBlue™ software, mouse, waste box, 100–240 V ±10 % / 50–60 Hz ±5 %, 0.2 µL–1 mL		5075000304