



NucleoSpin® 96 DNA RapidLyse

Automated isolation of total DNA from tissue and organs on the flowbot® ONE workstation

Application benefits

Experience enhanced nucleic acid purification workflows with the NucleoSpin® 96 DNA RapidLyse kit and flowbot® ONE, featuring:

- Verified methods ensuring a semi automated purification process.
- Consistent recovery of total DNA with reliable reproducibility in both yield and purity.
- High throughput capability, processing up to 96 samples in parallel.
- Accessible protocols through MACHEREY-NAGEL's technical automation support at automation-bio@mn-net.com.

Keywords

Total DNA, Nucleic acid extraction, Automated DNA purification, challenging samples, quick lysis, vacuum processing, flowbot® ONE, Flowrobotics automation system, laboratory protocol optimization



Flow Robotics flowbot® ONE

The flowbot® ONE Workstation was equipped with the NucleoVac 96 vacuum manifold an, external vacuum pump and digital vacuum regulator

Introduction

The isolation of genomic DNA from tissues, cells or challenging sample materials such as insects can be a time-consuming task. However, efficient lysis and DNA release is essential for subsequent downstream applications in molecular biological research. MACHEREY-NAGEL designed the silica membrane based NucleoSpin® 96 DNA RapidLyse kit with a unique buffer chemistry to significantly reduce the overall extraction time and cost compared to traditional methods. Depending on the sample material lysis can be performed on the automation platform in 15–60 min or for challenging samples the kit is also compatible with an external mechanical disruption and lysate clearing step using MN Bead tubes or Bead Plates.

High quality DNA can be extracted on flowbot® ONE and directly used as a template for qPCR, NGS, blotting, or various other enzymatic reactions. This silica membrane-based kit can be manually processed via centrifugation or automated by using a vacuum manifold. This application note describes the automated process on the automated liquid handling workstation flowbot® ONE using the NucleoSpin® 96 DNA RapidLyse kit from MACHEREY-NAGEL. The processing of 96 samples takes approximately 70 minutes excluding sample lysis.

NucleoSpin® 96 DNA RapidLyse

Technology	Silica membrane
Sample material	Bacteria, human or animal tissue, cells
Target molecules	total DNA
Fragment size	~200 bp – approx. 50 kbp
Sample numbers on flowbot® ONE	96 samples with 1000 µL 8-channel pipette configuration

flowbot® ONE

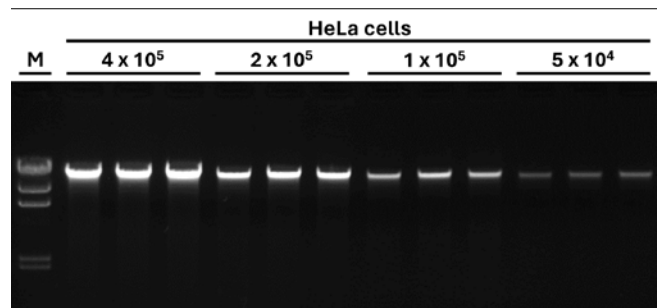
Technology	Automated liquid handling platform equipped with electronic pipettes
Sample numbers	1 – 96 samples
Deck positions	Configurable platform with 12 deck slots + tip waste
Pipetting volume	2 pipette modules (choose between 1, 4 and 8-channel) Volume ranges: 1 – 20 µL, 2 – 200 µL and 10 – 1000 µL

Material and Methods

The optimized flowbot® ONE protocol is programmed to process 96 samples in parallel. Cell culture samples were quickly lysed via a syringe needle, whole insects were disrupted via a swing mill using steel beads (e.g. MN Bead Tube Type D) and mouse samples (30 mg tissue) were lysed enzymatically in 40–60 minutes incubation agitated at 56 °C. The efficient release of DNA is facilitated by a meticulously designed lysing system with optimized parameters, incorporating the specialized Lysis Buffer RLY along with Liquid Proteinase K. Extended overnight or incubation for multiple hours is not necessary. During the binding phase, nucleic acids attach reversibly to the silica membrane. Subsequently, contaminants like salts or lipids are eliminated from the silica membrane through three washing stages utilizing Washing Buffer RLW, while nucleic acids remain bound. Ultimately, highly pure genomic DNA is extracted under low ionic strength conditions using the mildly alkaline Elution Buffer RLE.

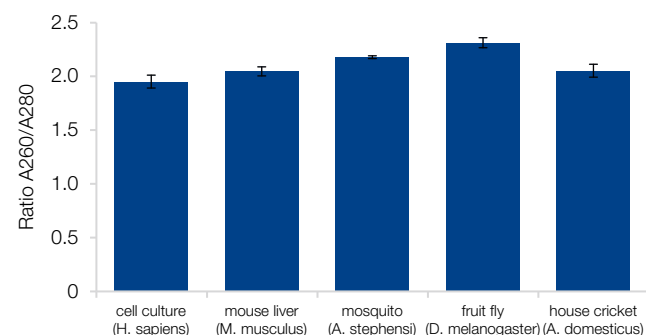
Application Data

Speed up and automate your gDNA extraction from e.g., tissue samples and cells. MACHEREY-NAGEL and Flow Robotics deliver a semi automated solution for your high throughput DNA extraction from various sample types. Quick lysis followed by a fast automated DNA extraction using the NucleoSpin® 96 DNA RapidLyse and fast processing of 96 samples in parallel.



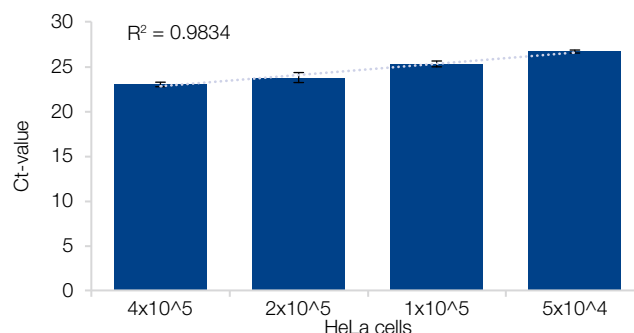
DNA extraction from different input amount of HeLa cell culture

DNA quality and integrity was visualized via gel electrophoresis (10 µL per lane, 1% TAE-gel), revealing consistently high-molecular DNA with a low degree of fragmentation. High quality DNA was extracted from various cell counts of HeLa cells using the NucleoSpin® 96 DNA RapidLyse kit on the flowbot® ONE system.



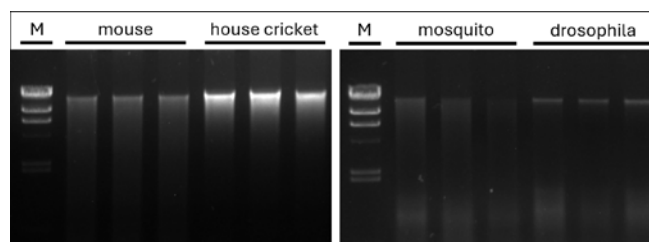
Purity of isolated DNA from different starting materials

DNA extraction was conducted using the NucleoSpin® 96 DNA RapidLyse kit on the flowbot® ONE system. DNA purity was determined photometrically. The results show consistent high purity Ratio A260/A280 determined from DNA isolated 4 × 10⁵ HeLa Cells, 30 mg Mouse liver and different insect samples like, Mosquitos (10 per extraction), Drosophila (5 per extraction) and domestic house crickets (1 per extraction).



Reliable qPCR of isolated DNA from a 1:2 dilution series of HeLa cells

qPCR targeted the mammalian beta-Actin gene and was conducted using the SensiFast™ Probe Lo-ROX kit from BioLine on an Applied Biosystems® 7500 Real-Time PCR System. Results show a sensitive detection and amplification across all different starting amounts and a high degree of integrity indicated via the linear regression with an R²-value of 0.98.



DNA extracted from different starting materials

DNA extraction was conducted from mouse tissue lysed enzymatically and whole insects lysed mechanically via bead beating using the NucleoSpin® 96 DNA RapidLyse kit on the flowbot® ONE system. DNA quality was visualized via gel electrophoresis (~500 ng per Lane, 1% TAE-gel), showing low degree degradation and high molecular DNA bands.

Ordering information

Product	Specifications	Pack of	REF
NucleoSpin® 96 DNA RapidLyse	Rapid extraction of total DNA from tissue and organs in proven 96-well plate format; including NucleoSpin® 96 DNA RapidLyse Binding Plate, reagents, Proteinase K and plastic consumables (MN Wash Plate, Square-well Block) (only elution plate required)	1 × 96 preps	740110.1
		4 × 96 preps	740110.4
Flow Robotics Flowbot® ONE Workstation	Automated platform for automated liquid handling and sample preparation	1 – 96 samples	

NucleoSpin® is a registered trademark of MACHEREY-NAGEL (contact: automation-bio@mn-net.com); Flowbot® is a registered trademark of Flow Robotics; Applied Biosystems® is a registered trademark of Applied Biosystems.

*For more detailed information, please visit www.flow-robotics.com. To contact Flow Robotics Sales or to schedule a demo, please email info@flow-robotics.com.