NucleoSpin® 96 Plant II

Automated isolation of total DNA from diverse plant samples on the flowbot® ONE workstation

Application benefits

Experience enhanced nucleic acid purification workflows with the NucleoSpin® 96 Plant II 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, inhibitor removal, plant, agriculture, phylogenetics, GMO testing, vacuum, 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

DNA extraction from plants is essential in various research fields, including phylogenetics, biodiversity studies, agricultural crop improvement, disease resistance, plant breeding, genetic engineering or even medical plant research. Techniques like CRISPR/Cas9, TILLING, SSR, and QTL mapping require reliable high-throughput DNA extraction. Common issues include low DNA quality, limited sample throughput and the release of plant metabolites like polyphenolic compounds or complex polysaccharides . This application note presents a solution using the MACHEREY-NAGEL NucleoSpin® 96 Plant II kit with the FlowRobotics flowbot® ONE workstation. The described automated pipetting system ensures reproducibility in yield and quality while minimizing manual interactions through automated liquid handling and vacuum filtration, extracts pure DNA from up to 96 samples in 70 minutes. The NucleoSpin® 96 Plant II kit achieves excellent DNA quality and yield, suitable for various downstream applications.

Accurate and efficient DNA extraction is vital for these research areas to obtain high-quality genetic material for further analysis

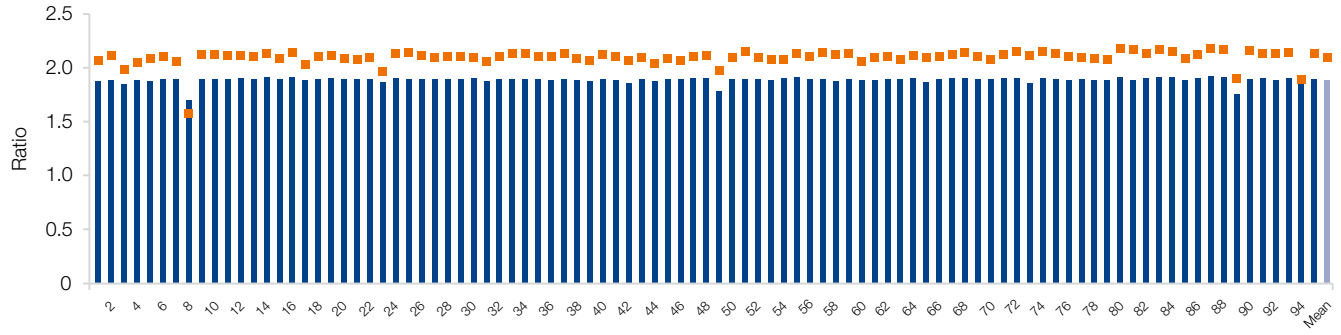
NucleoSpin® 96 Plant II	
Technology	Silica membrane
Sample material	Plant tissue
Target molecules	Total DNA
Fragment size	50 bp – approx. 50 kbp
Sample numbers on flowbot® ONE	96 samples with 1000 µL 8-channel pipette configuration

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

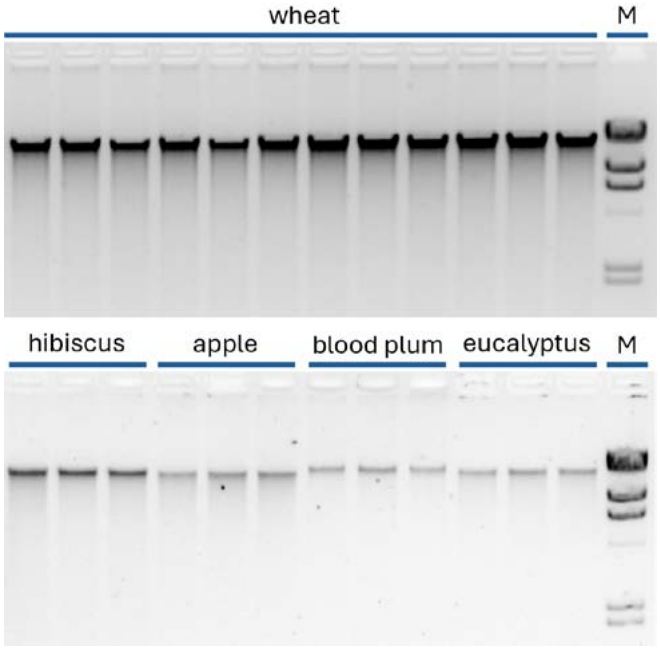
The NucleoSpin® 96 Plant II kit is used for isolating DNA from various plant species. Up to 100 mg of homogenized plant material (e. g., leaf, root) is processed. The kit contains two optional lysis systems for efficient lysis of a wide range of plants. The Lysis Buffer PL1, containing CTAB, effectively removes polysaccharides. Alternatively, the SDS based lysis buffer PL2 can be used to precipitate proteins under the addition buffer PL3. Cleared lysate is mixed with Binding Buffer PC to create optimal binding conditions. The nucleic acid is reversible to the silica membrane during the first vacuum filtration step. Subsequent wash steps with different wash buffers (PW1 and PW2) remove contaminants before the DNA is eluted under low ionic conditions by using the Elution buffer PE.

Application Data



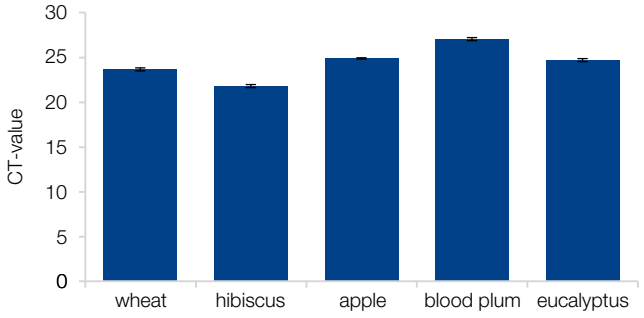
Reliable and reproducible purity across a full plate of 96 extractions

DNA extraction was conducted from 20 mg wheat leaves per sample using the NucleoSpin® 96 Plant II kit on the flowbot® ONE system. DNA purity was determined photometrically Ratio A_{260}/A_{280} (blue bars) and Ratio A_{260}/A_{230} (orange boxes). Results show consistently high purity for both ratios across the entire plate.



DNA extraction from five different plant samples

High DNA quality and integrity was visualized via gel electrophoresis (1 % TAE-gel), revealing consistently high-molecular DNA. The top (A) shows 12 randomized selected wheat (Triticum aestrivum) DNA samples from a full 96 sample extraction run, showing an even yield across the entire plate. Different lysis conditions are shown below (B) with hibiscus (Hibiscus syriacus) and apple (Malus domestica) leaves being lysed in Buffer PL1. Blood plum (Prunus cerasifera) and eucalyptus (Eukalyptus spec.) leaves were lysed with the combination of Buffers PL2 and PL3. These results demonstrate the efficacy of the NucleoSpin® 96 Plant II kit and the flowbot® ONE system in achieving reliable and high-quality DNA isolations from a variety of plant samples.



qPCR amplification of DNA extracted from various plant types

DNA extraction was conducted from leaf material of wheat, hibiscus, apple, blood plum and eucalyptus using the NucleoSpin® 96 Plant II kit on the flowbot® ONE system. Subsequent qPCR analysis, targeting a highly conserved photosynthetic relevant part of the chloroplast genome, was conducted using the SensiFast™ probe Lo-ROX kit from BioLine on an Applied Biosystems® 7500 Real-Time PCR System. Results show a high amplification of DNA across all sample materials, indicating the efficient removal of inhibitors and bias free lysis of plant samples (n= 3 each).

Ordering information

Product	Specifications	Quantity	REF
NucleoSpin® 96 Plant II	Rapid isolation of DNA from a multitude of plant samples in proven 96-well plate format; including NucleoSpin® Plant II Binding Plate, buffers and plastic consumables (MN Wash Plate, and Rack of tube Strips for elution)	2 × 96 preps 4 × 96 preps 24 × 96 preps	740663.2 740663.4 740663.24
Square-well Block	96 Deep-well Plate to provide cleared lysates	4 pieces 24 pieces	740481 740481.24
Flow Robotics flowbot® ONE	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 FlowRobotics; Applied Biosystems® is a registered trademark of Applied Biosystems.
*For more detailed information, please visit www. flow-robotics.com. To contact FlowRobotics Sales or to schedule a demo, please email info@flow-robotics.com.