MACHEREY-NAGEL

Application Note 03/2025 · Bioanalysis

NucleoMag® HMW DNA

Automated purification of high molecular weight (HMW) DNA from plants on the MagnetaPure 32+

Application benefits

The integration of the proven NucleoMag® HMW DNA technology and the MagnetaPure 32+ provide a streamlined and efficient workflow for isolating high-quality high molecular weight DNA from diverse plant samples, enabling the technical challenges of sample preparation for long-read sequencing and facilitating efficient genome assembly:

- Verified automation method for isolating high molecular weight DNA in plant studies
- Simultaneous processing of up to 32 samples in parallel
- Consistent and reliable results
- No programming required: Verified and pre-installed methods available

Keywords

High molecular weight (HMW) DNA, Oxford Nanopore Technologies, Pacific Biosciences, plants, NucleoMag®, magnetic beads, magnetic rod system, MagnetaPure

Introduction

The extraction of high molecular weight (HMW) DNA from plants presents significant technical challenges, especially when preparing samples for long-read sequencing technologies, such as those offered by Oxford Nanopore Technologies or Pacific Biosciences. These technologies require long, intact DNA molecules to achieve accurate and comprehensive genome assembly. However, obtaining HMW DNA from plants is complicated due to several factors inherent to plant biology.

Plants have complex cell walls rich in polysaccharides and lignins, making cell lysis more difficult compared to other organisms. In addition, plants contain secondary metabolites such as polyphenols and tannins, which can co-precipitate with DNA, leading to contamination and degradation of the sample. These contaminants not only interfere with downstream processing, but can also shear the DNA, reducing its length and quality, which is critical for long-read sequencing.

The NucleoMag® HMW DNA kit is designed to extract high molecular weight DNA from a variety of samples including plants. This kit provides a reliable solution for obtaining high quality and high purity DNA suitable for downstream genomic analysis.

This application note highlights the automated extraction of DNA using the NucleoMag® HMW DNA kit and the MagnetaPure 32+ system. This compact nucleic acid extraction platform can process up to 32 samples simultaneously in 35 to 45 minutes. It simplifies the workflow by automating the mixing, magnetic bead transfer, washing, and elution steps.

For complicated plant species, we performed an additional purification step with NucleoMag® NGS Clean-up and Size Select using the MagnetaPure 32+. This method improves the recovery of samples with low OD ratios without compromising the DNA integrity.

NucleoMag® HMW DNA	
Technology	Magnetic beads
Sample material	≤ 50 mg plant leaves
Elution volume	100 – 200 µL
Fragment size	≥ 50 kb – Up to 100 – 200 kb
Max. sample number on the MagnetaPure 32+	32 samples

www.mn-net.com/nucleomag-hmw-dna-high-molecular-weight-dna-from-diverse-sample-materials-744160.1

MagnetaPure 32+	
Technology	Automated magnetic rod system
Display	7 inch-color touch screen
Capacity / volume per well	1 – 32 samples / 30 µL to 1000 µL
Dimensions	417 x 410 x 426 mm
Weight	30 kg
Contamination control	UV lamp, internal filter system
Website	*www.mn-net.com/MagnetaPure32*

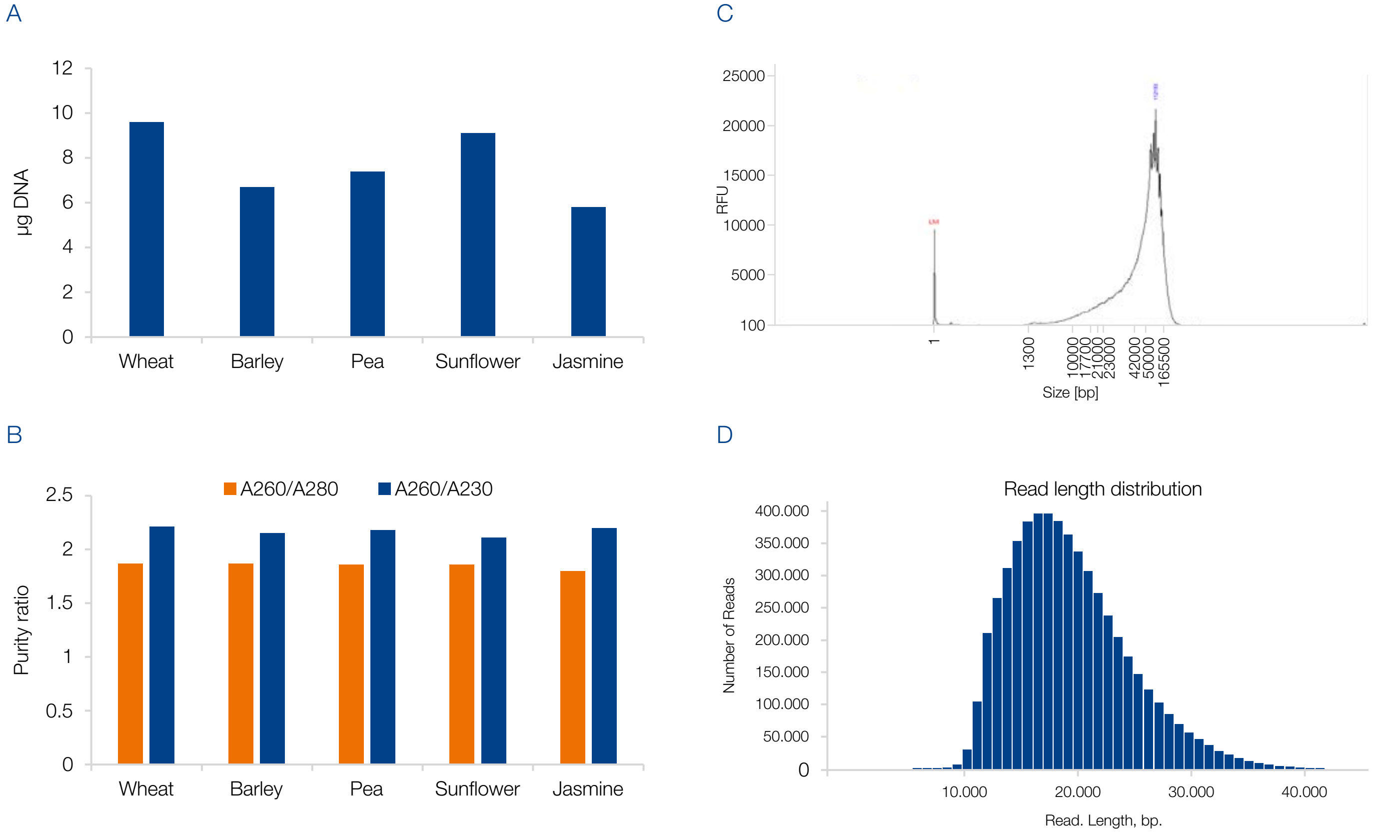
Material and Methods

The isolation procedure of the NucleoMag® HMW DNA Kit is based on the reversible adsorption of nucleic acids onto paramagnetic NucleoMag® B-Beads under appropriate binding conditions. For sample preparation, 50 mg of frozen plant tissues (young leaves) were disrupted by bead beating in a 2 mL Eppendorf tube (Two 5 mm steel beads with Retsch® disrupter at 30s at 20Hz). Lysis and RNase digestion were performed off deck according to the NucleoMag® HMW DNA user manual. Lysates were transferred to a 96 Deep-well plate prefilled with NucleoMag® B-Beads and Binding Buffer (HM2), Wash Buffers (HM3, HM4, EtOH 70%) and Elution Buffer (HM6). All steps involving binding, washing and magnetic bead separation were performed using the MagnetaPure 32+ magnetic rod system.

The integrity and quality of the extracted DNA samples were verified and were within the recommendations for long read sequencing. Most of the samples presented in this application note were successfully sequenced on PacBio Revio or ONT P2 Solo systems.

Long-read sequencing enables the sequencing of several kilobases DNA fragments. This method is particularly useful for resolving complex regions of the genome, such as repetitive sequences or structural variations. PacBio and Oxford Nanopore Technologies (ONT) offer long-read sequencing technologies. The DNA isolated from rose bush and sunflower samples with the NucleoMag® HMW DNA kit provided sufficient quality and quantity to enable high-resolution sequencing, ensuring reliable data respectively with ONT technology for rose bush DNA, and with PacBio technology for sunflower DNA.

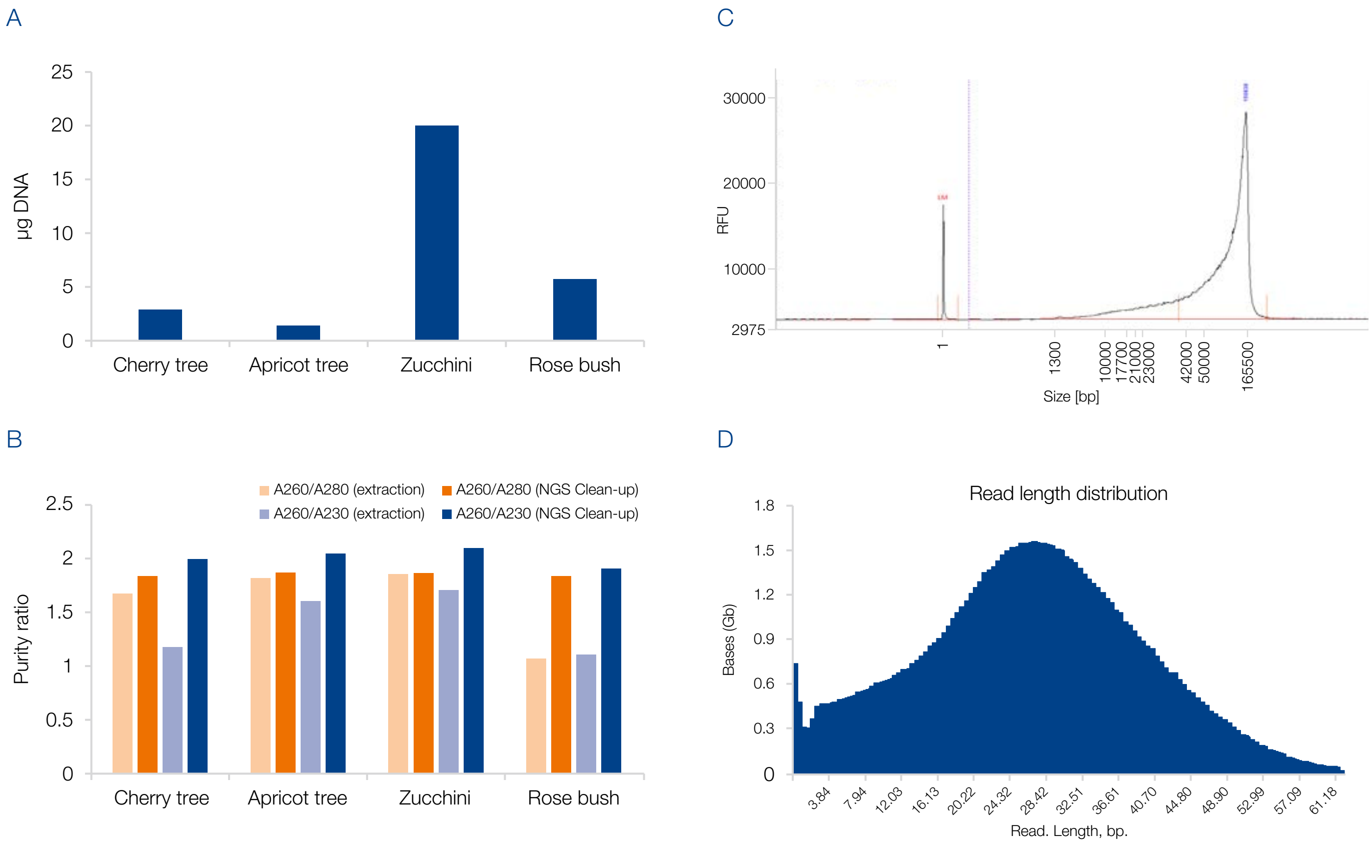
Application Data



High quality and high molecular weight DNA from diverse plant materials

DNA of various plant species (Wheat, Barley, Pea, Sunflower, Jasmine) were extracted with NucleoMag® HMW DNA procedure using MagnetaPure 32+ system. Nucleic acids were quantified by fluorescence (Qubit fluorometer – Invitrogen) (A). Purity was determined by UV spectroscopy (B). Integrity of HMW DNA was assessed using the Femto Pulse system (Agilent) with the Genomic DNA 165 kb Kit. An example of the data obtained was given for the sunflower (C). Sunflower sequencing data on 1 SMRT cell 25M on a PacBio Revio System (D).

	Sunflower (PacBio sequencing)
Yield	105,6 Gb
Number of reads	5,5 Millions
N50	19 Kb



High quality and high molecular weight DNA from complex plant materials

Metrics of some DNA samples from difficult species (Cherry tree, Apricot tree, Zucchini, Rose bush) didn't meet the purity specifications for downstream analysis. DNA from these complex plant species was extracted using the NucleoMag® HMW DNA procedure on the MagnetaPure 32+ system. The elution from each sample was then purified using the NucleoMag® NGS Clean-up and Size Select Kit on the MagnetaPure 32+ to achieve purity for downstream applications. Nucleic acids were quantified by fluorescence (Qubit fluorometer – Invitrogen) (A). Purity was determined by UV spectroscopy (B). Integrity of HMW DNA was assessed using the Femto Pulse system (Agilent) with the Genomic DNA 165 kb Kit. An example of the data obtained was given for the Rose bush (C). Rose bush sequencing data on 1 flowcell on a P2 Solo ONT System (D).

	Rose bush (ONT sequencing)
Yield	93,6 Gb
Number of reads	8,5 Millions
N50	27 Kb

Ordering information

Product	Specifications	Quantity	REF
NucleoMag® HMW DNA	Magnetic bead based kit for the isolation of high molecular weight DNA from plant samples; including NucleoMag® B-Beads and buffers	1 x96 preps	744160.1
MagnetaPure 32+	Magnetic rod system for automated nucleic acid extraction using MACHEREY-NAGEL NucleoMag® kits, parallel processing of up to 32 samples	1	747010
NucleoMag® NGS Clean-up and Size Select	Magnetic bead-based kit for clean-up and size selection of Next Generation Sequencing library preparation reactions	5 mL 50 mL 500 mL	744970.5 744970.50 744970.500
96 Deep-well plates	96 Deep-well plates for MagnetaPure 32+	25	744955
Tip combs	8-place magnetic tip combs for MagnetaPure 32+	50	744960

Learn more

To learn more about the French National Resources Centre for Plant Genomics (CNRGV), visit the website *https://cnrgv.toulouse.inrae.fr/*, or contact isabelle.dufau@inrae.fr, anthony.theron@inrae.fr, infocnrgv-toulouse@inrae.fr.

To learn more about MACHEREY-NAGEL's NucleoMag® HMW DNA and MagnetaPure 32+ purification solutions, please visit *www.mn-net.com*.

Acknowledgements

This protocol was developed by MACHEREY-NAGEL application scientists and is intended for research use only. Users are responsible for determining the suitability of the protocol for their application.

About the Authors

Isabelle Dufau and Anthony Theron are biology engineers at French National Resources Centre for Plant Genomics (CNRGV). The CNRGV is a national infrastructure belonging to the French National Research Institute for Agriculture, food and Environment (INRAE). Settled in Toulouse (France) in 2004, the CNRGV is both a Biological Resources Centre (BRC) dedicated to all plant genomic resources and a service provider for plant genomics projects.

It aims at providing innovative and efficient genomic tools to better characterize plant biodiversity and understand how plants adapt to their environment through the analysis of their genomes.

Its field of expertise includes the development of HMW DNA extraction protocols dedicated to plants, targeted sequencing of genomic regions of interest and plant whole genome characterization “From seed to the genome” (genomic sequences produced in collaboration with sequencing facilities). The CNRGV has also developed bioinformatics expertise to analyse and explore the complex structures of plant genomes.

Olivier Foumenaigue is a senior application specialist in molecular biology automation at MACHEREY-NAGEL. He holds a Master's degree in Biotechnology and Biological Engineering from the Université de Technologie de Compiègne. With a background in forensic science and over a decade of experience in high throughput DNA extraction, automation and customer support, he has held key roles in both field applications and service leadership across Europe. Since joining MACHEREY-NAGEL in 2021, his work has focused on advancing genomic workflows and optimising automation strategies for nucleic acid purification.

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