



GENETIX BRAND

HANDBOOK



Nucleopore FFPE DNA Miniprep Kit

Nucleopore® FFPE DNA Miniprep Kit

GX-4076

50 Prep



Genetix Biotech Asia Pvt. Ltd.

71/1, First Floor, Shivaji Marg, Najafgarh Road, New Delhi - 110015

Phone : +91-11-45027000 ■ Fax : +91-11-25419631

E-mail : info@genetixbiotech.com ■ www.genetixbiotech.com



www.genetixbiotech.com

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COMPONENTS

Kit contents

Nucleopore FFPE DNA MiniPrep™

Cat. No. (Kit Size)	GX-4076 (50 Preps.)	Storage
Proteinase K & Storage Buffer ¹	2 x 5 mg	20°C (after mixing)
2X Digestion Buffer	5 ml	Room Temp.
Genomic Lysis Buffer ²	50 ml	Room Temp.
DNA Pre-Wash Buffer	15 ml	Room Temp.
g-DNA Wash Buffer	50 ml	Room Temp.
DNA Elution Buffer	10 ml	Room Temp.
G-Spin Columns	50	Room Temp.
RNase A ³	2 x 1 mg	4°C
Collection Tubes	100	Room Temp.
Instruction Manual	1	-

Note - Integrity of kit components is guaranteed for up to one year from date of purchase. Reagents are routinely tested on a lot-to-lot basis to ensure they provide the highest performance and reliability.

1 The Proteinase K is stable as shipped. Add 260 µl Proteinase K Storage Buffer to each Proteinase K tube prior to use. The final concentration of Proteinase K after the addition of Proteinase K Storage Buffer is ~20 mg/ml. Store at -20° C.

2 Recommended: Add beta-mercaptoethanol to 0.5%(v/v) i.e., 250 µl per 50 ml or 500 µl per 100 ml.

3 Re-suspend lyophilized RNase A in 150 µl per tube of ddH₂O. Store at 4° C.

Specification

- Sample Size – Up to 25 mg tissue from paraffin block or up to four (4) tissue sections (≤20 µm thick) with a total surface area ~20 cm². It is recommended to use 1-2 sections if performing the protocol for the first time. Compatible with fresh/frozen tissue specimens.
- DNA Recovery – The maximum DNA binding capacity of the provided spin column is ~25 µg.
- Processing Time – Not provided with the Kit
- Equipment/Reagents – Microcentrifuge, thermomixer or heat block/bath capable of 55°C and 90°C, xylene, ethanol, isopropanol, beta-mercaptoethanol (optional).

Product Description

The Nucleopore FFPE DNA MiniPrep™ provides a simple and reliable method for high yield/quality DNA isolation from formalin-fixed, paraffin embedded (FFPE) tissue samples and sections. The unique chemistries of the product have been optimized for maximum recovery of non-crosslinked, ultra-pure DNA without RNA contamination. Simply digest deparaffinized tissues using the provided Proteinase K, heat, and then purify the DNA with the Fast-Spin columns in the kit. PCR inhibitors are effectively removed during the isolation procedure, and eluted DNA is ideal for PCR, Next-Gen library prep, enzymatic manipulation, etc.

Highlights

- High performance sample prep technology for high quality DNA (up to ~25 µg/prep) from FFPE tissue samples and sections.
- Selectable size cut-off technology; recover total DNA > 50 bp or > 500 bp.
- Eluted DNA is RNA-free and ideal for PCR, Next-Gen library prep, enzymatic manipulation, etc.

Buffer preparation

Add 260 µl Proteinase K Storage Buffer to each Proteinase K tube prior to use and store at -20° C. The final concentration of Proteinase K after the addition of Proteinase K Storage Buffer is ~20 mg/ml.

Resuspend lyophilized RNase A in 150µl per tube of ddH₂O. Store at 4° C.

Recommended: Add beta-mercaptoethanol (user supplied) to the Genomic Lysis Buffer to a final dilution of 0.5%(v/v) i.e., 250 µl per 50 ml.

Protocol

Rapid Deparaffinization (Slide Tissue Sections Only)

1. Remove (trim) excess paraffin wax from sample and transfer the sample to a 1.5 ml microcentrifuge tube.
2. Add 1 ml xylene (not provided) to the sample. Vortex vigorously for 30 seconds and then centrifuge sample at 10,000 x g (~10,000 rpm) for 1 minute. Remove and discard the xylene.
3. Wash sample with 1 ml ethanol (95-100%). Vortex vigorously for 30 seconds then centrifuge samples at 10,000 x g for 1 minute. Remove and discard ethanol. Repeat this step.
4. Dry the sample using vacuum centrifugation (e.g., SpeedVac or similar) or by heating uncapped tubes at ~37° C for up to 40 minutes.
5. The sample is now ready for Proteinase K Digestion & DNA Isolation.

Standard Deparaffinization (Tissue Samples and Slide Tissue Sections)

1. Remove (trim) excess paraffin wax from sample and transfer the sample to a 1.5 ml microcentrifuge tube.
2. Add 1 ml xylene (not provided) to the sample. Vortex and incubate at room temperature for 1 hour with gentle rocking. Centrifuge, discard supernatant, and repeat this step.
Note: Centrifuge at 10,000 x g for 1 minute and remove/discard supernatant after washing for the following steps.
3. Wash twice with 1 ml ethanol (100%) for 5 minutes with gentle rocking.
4. Wash twice with 1 ml ethanol (95%) for 5 minutes with gentle rocking.
5. Wash twice with 1 ml ethanol (75%) for 5 minutes with gentle rocking.
6. Wash once with 1 ml ddH₂O for 5 minutes with gentle rocking. Remove as much water from the sample as possible.
7. The sample is now ready for Proteinase K Digestion and DNA Isolation

Proteinase K Digestion & DNA Isolation

1. To a deparaffinized tissue sample (≤25 mg) in a microcentrifuge tube, add the following:

H ₂ O	45µl
2X Digestion Buffer	45µl
Proteinase K	10µl

Note: If your tissue sample is too large for the digestion volume, scale up the digestion to 200 µl while keeping the amount of Proteinase K the same. Double the reagent volumes indicated in Step 3 & 4.

2. Rapid Digestion

Incubate at 55°C for 1-4 hours

Standard Digestion

Incubate at 55°C overnight (12-16 hrs)

Note: The Rapid Digestion is recommended for processing slide tissue sections but should also be sufficient for most tissue samples. However, the Standard Digestion ensures maximum yields of DNA from even tough-to-lyse (collagen-rich, fibrous, etc.) tissue samples.

3. Transfer the digestion to 94°C and incubate for 20 minutes. When done add 5 µl of RNase A, mix, and then incubate an additional 5 minutes at room temperature.

Note: It is recommended to skip the heat treatment in Step 3 and the addition of isopropanol in Step 4 if only double stranded DNA is required.

4. Add 350 µl of Genomic Lysis Buffer to the tube and mix thoroughly by vortexing.

To Isolate Total DNA >50 bp

Add 135 µl of isopropanol* (user supplied) to the sample, mix thoroughly, and proceed to Step 5

To Isolate DNA >500 bp

Proceed directly to Step 5

Note: When working with a new sample, it is recommended to isolate total DNA as a precaution. FFPE DNA may be highly degraded and DNA > 500 bp may not be present in sample.

5. Centrifuge at 10,000 x g for 1 minute to remove insoluble debris and then transfer the supernatant to a G-spin™ Column in a Collection Tube. Centrifuge at 10,000 x g for 1 minute.
6. Add 200 µl of DNA Pre-Wash Buffer to the spin column in a new Collection Tube. Centrifuge at 10,000 x g for 1 minute.
7. Add 400 µl of g-DNA Wash Buffer to the spin column. Centrifuge at 10,000x g for 1 minute.
8. Transfer the G -Spin™ Column to a clean microcentrifuge tube. Add ≥50 µl DNA Elution Buffer or water (e.g., add ≥100 µl if sampling 25 mg tissue) to the spin column. Incubate 2-5 minutes at room temperature, then centrifuge at top speed for 30 seconds to elute the DNA. The eluted DNA can be used immediately for molecular based applications or stored ≤-20°C for future use.

ORDERING INFORMATION

Description	Pack Size	Cat. No.
* DNASure Tissue Mini Kit	50 preps	NP-61305
* DNASure Plant Mini Kit	50 preps	NP-79105
* DNASure Plant Mini Kit	250 preps	NP-79107
* DNASure Plant Midi Kit	20 preps	NP-78153
* DNASure Plant Maxi Kit	10 preps	NP-78164
* DNASure Blood Mini Kit	50 preps	NP-61105
* DNASure Blood Mini Kit	250 preps	NP-61107
* DNASure Blood Midi Kit	20 preps	NP-61184
* DNASure Blood Maxi Kit	10 preps	NP-61193
* DNASure Blood FastPure Kit	50 preps	NP-62205
* DNASure Blood FastPure Kit	250 preps	NP-62207
* SureSpin Plasmid Mini Kit	50 preps	NP-37105
* SureSpin Plasmid Mini Kit	250 preps	NP-37107
* SureSpin Plasmid FastPrep Kit	50 preps	NP-47105
* SureSpin Plasmid FastPrep Kit	250 preps	NP-47107
* SureSpin Buffer Set*	1	37107-BS
* SurePrep Plasmid Mini Kit	20 preps	NP-15123
* SurePrep Plasmid Mini Kit	100 preps	NP-15125
* SurePrep Plasmid Midi Kit	20 preps	NP-15143
* SurePrep Plasmid Midi Kit	100 preps	NP-15145
* SurePrep Plasmid Maxi Kit	10 preps	NP-15161
* SurePrep Plasmid Maxi Kit	25 preps	NP-15162
* SurePrep Plasmid Mega Kit	5 preps	NP-15183
* SurePrep Plasmid Giga Kit	5 preps	NP-15191

*SureSpin® Buffer Set

For the isolation of low-copy plasmids, buffers PA1, PA2, PA3, RNase A, sufficient for 300 preps

ORDERING INFORMATION

Description	Pack Size	Cat. No.
SurePrep® Buffer Set**	1	15143-BS
SurePrep® Plasmid Endofree Maxi Kit	10 preps	NP-15363
SurePrep Plasmid Endofree Mega Kit	5 preps	NP-15365
SurePrep® Plasmid Endofree Giga Kit	5 preps	NP-15367
SureSpin® 96 PCR Kit	4x96	NP-38151
SureTrap® Gel Extraction Kit	50 preps	NP-38705
SureTrap® Gel Extraction Kit	250 preps	NP-38707
SureTrap® PCR Cleanup Kit	50 preps	NP-38105
SureTrap® PCR Cleanup Kit	250 preps	NP-38107
SureExtract® Spin PCR/Gel Extraction Kit	50 preps	NP-36105
SureExtract® Spin PCR/Gel Extraction Kit	250 preps	NP-36107
SureSEQ® Cleanup Kit	50 preps	NP-73205
RNASure® Mini Kit	50 preps	NP-84105
RNASure® Mini Kit	250 preps	NP-84107
RNASure® Plant Kit	50 preps	NP-84905
RNASure® Plant Kit	250 preps	NP-84907
miRNASure® Mini Kit	50 preps	NP-71002
SureTrap® mRNA Mini Kit	12 preps	NP-80033
SureTrap® mRNA Midi Kit	12 preps	NP-80043
RNASure® Virus Kit	50 preps	NP-67705
RNASure® Virus Kit	250 preps	NP-67707

**SureSpin® Buffer Set

For isolation of low-copy plasmids, cosmids, BACs, PACs, and P1 constructs, only applicable with SurePrep® Plasmid kits, sufficient for 10 SurePrep Maxi Columns (Maxi preps), 20 SurePrep® Midi Columns (Midi preps), set incl. RNase A

ORDERING INFORMATION

Description	Pack Size	Cat. No.
Nucleo-pore® Stool DNA Mini Kit	50	NP-7011D
Nucleo-pore® gRNA Blood Kit	50	NP-0201R
Nucleo-pore® gDNA Urine Kit	20	NP-6030D
Nucleo-pore® Yeast Transformation Kit	120	NP-1002T
Nucleo-pore® DNA Methylation Kit	50	NP-6006D
Nucleo-pore® gDNA Clean-up Kit	200	NP-4304D
Nucleo-pore® Bisulphite DNA Clean-up Kit	50	NP-5205D
Nucleo-pore® gDNA Fungal/Bacterial Mini Kit	50	NP-7006D

Product Warranty

Nucleopore® FFPE DNA Miniprep Kit components are intended for research purposes only. They are suitable for *in vitro* uses only. The purchaser must determine the suitability of the product for its particular use. Should any product fail to perform satisfactorily due to any reason other than misuse, Genetix will replace it free of charge or refund the purchase price. Genetix reserve the right to change, alter, or modify any product to enhance its performance and design. It is the responsibility of the user to verify the use of the Nucleopore® FFPE DNA Miniprep Kit for a specific application range as the performance characteristic of this kit has not been verified to a specific organism. No claim or representation is intended for its use to identify any specific organism or for clinical or therapeutic use.

Genetix does not warrant against damages or defects arising in shipping and handling (transport insurance for customers excluded), or out of accident or improper or abnormal use of this product.

In accordance with Genetix ISO-certified Quality Management System, each lot of Nucleopore® FFPE DNA Miniprep Kit is tested against predetermined specifications to ensure consistent product quality.

In no event shall Genetix be liable for claims for any other damages, whether direct, indirect, incidental, compensatory, foreseeable, consequential, or special (including but not limited to loss of use, revenue or profit), whether based upon warranty, contract, tort (including negligence) or strict liability arising in connection with the sale or the failure of Genetix products to perform in accordance with the stated specifications.

Product claims are subject to change. Therefore please contact our Technical Support Department for updated information on Genetix products.

Please contact:

Genetix Biotech Asia (P) Ltd.

71/1, Najafgarh Road, Shivaji Marg,

New Delhi. 110015.

INDIA.

E-mail: info@genetixbiotech.com

techsupport@genetixbiotech.com

Tel: +91-11-45027000

Fax: +91-11-25419631