



GENETIX BRAND

HANDBOOK



Nucleopore[®] GDNA Cleanup Kit

- | | | | |
|--------------------------|--|-----------|-----------|
| <input type="checkbox"/> | Nucleopore [®] GDNA Cleanup Kit | NP- 3304D | 50 Preps |
| <input type="checkbox"/> | Nucleopore [®] GDNA Cleanup Kit | NP- 4304D | 200 Preps |



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COMPONENTS

Kit contents

Nucleopore® GDNA Cleanup Kit

| Cat # | NP-3304D | NP-4304D |
|--------------------|-------------|--------------|
| No. of Preps | (50 preps.) | (200 preps.) |
| Binding Buffer CCB | 50ml | 200ml |
| Wash Buffer CCW* | 6ml | 24ml |
| Elution Buffer CCE | 4ml | 10ml |
| Spin Columns | 50 | 200 |
| Collection Tube | 50 | 200 |
| Handbook | 1 | 1 |

*Please see "Preparation and Storage of reagents"

Reagents, consumables and equipments not provided with the kit

- Ethanol
- Pipette and Tips
- Centrifuge
- Vortexer

SAFETY INSTRUCTIONS

When working with chemicals, always wear a suitable labcoat, disposable gloves and protective goggles. For more information, please consult the appropriate material safety data sheets (MSDS). DNA Binding Buffer CCB contain guanidine salts, which can form reactive compounds when combined with bleach, in case of any spillage of buffer always use suitable laboratory detergent and water.

Binding Buffer CCB

Contain Guanidine Hydrochloride: R&S Phrases: R10-22-36/38, S7-16

R10: Flammable, R22 Harmful by Inhalation, in contact with skin and if swallowed, R36/38: Irritating to eyes and skin. S7: Keep container tightly closed. S16: Keep away from sources of ignition- No Smoking.

INTRODUCTION

Nucleopore GDNA Cleanup Kit provides a convenient and user friendly method for the rapid purification and concentration of up to 25 µg of high-quality DNA from PCR, endonuclease digestions, or DNA from cell lysates.

Principle and Procedure

The kit utilizes specially formulated DNA Binding Buffer CCB combined with Spin column technology resulting in elution of high quality and pure DNA suitable for nucleotide sequencing, microarray analysis, PCR, nucleotide blotting, and restriction endonuclease digestion procedures.

Specifications of Nucleopore GDNA Cleanup Kit

DNA Purity – High-quality, purified DNA is eluted with water and is especially well suited for sequencing, ligation reactions, and restriction endonuclease digestions.

DNA from PCR, restriction endonuclease digestions, plasmid preparations, kinase reactions, etc can be purified using Nucleopore GDNA Cleanup and concentrator kit. DNA from 50 bp to 23 kb can be purified using this kit, resulting in typically, up to 25 µg total DNA (in 25 µl water). Recovery of 50 bp to 10 kb is 70-90% whereas, for DNA from 11 kb to 23 kb the recovery is 50-70%.

Preparation and Storage of reagents

Wash Buffer CCW

Add 24ml and 96ml of absolute (ethanol (or 26ml and 104ml of 95% ethanol) to 6ml and 24ml of Wash Buffer CCW Concentrate respectively. Kit components can be stored for up to one year from date of purchase.

PROTOCOL FOR CLEANUP OF DNA

Things to do before starting

Check whether Wash Buffer CCW has been reconstituted as per instructions

Procedure

1. **In a 1.5 ml microcentrifuge tube, add 2 volumes of Binding Buffer CCB to each volume of DNA sample. (e.g., 300 µl binding buffer to 150 µl DNA sample). Mix briefly by vortexing.**

Note: For ssDNA (e.g., single stranded cDNA), add 7 volumes to each volume of sample. For example, 350 µl Binding Buffer CCB to 50 µl sample.

2. **Transfer mixture to a provided Spin Column in a 2ml Collection Tube.** The sample capacity of the column is 800µl. Therefore, it may be necessary to load and spin a column multiple times if total volume is more than 800µl.
3. **Centrifuge at $\geq 10,000 \times g$ for 30 seconds. Discard the flow-through.**
4. **Add 200 µl Wash Buffer CCW to the column. Centrifuge at $\geq 10,000 \times g$ for 30 seconds.**
5. **Again add 200 µl Wash Buffer CCW to the column. Centrifuge at $\geq 10,000 \times g$ for 30 seconds.**
6. **Add 30 - 50 µl water or elution buffer directly to the column matrix. Transfer the column to a 1.5 ml microcentrifuge tube and centrifuge at $\geq 10,000 \times g$ for 30 seconds to elute the DNA.**

Note: Elution of DNA from the column is dependent on pH and temperature. If water is used, make sure the pH is > 5.0 . Waiting 1 minute after adding water to the column may improve the yield of larger (> 6 kb) DNA. For larger DNA (> 10 kb) the total yield may be improved by eluting the DNA with 60-70°C water. TE buffer (10 mM Tris-HCl, 1 mM EDTA, pH 8.0) or modified TE (10 mM Tris, 0.1 mM EDTA, pH 8.5) can also be used for elution if required by your experiment.

The ultra-pure DNA in water is now ready for use for downstream applications.

TROUBLESHOOTING GUIDE

Low Yield

Possible cause

- pH of elution buffer/water too low

Suggestions

- Check the pH of Elution Buffer (1XTE Buffer pH 8.0) or water used for elution. It should be more than 5. Pre-warm elution buffer/water before elution.

Possible cause

- Starting material too low

Suggestions

- Add 100 µl binding buffer CCB to all samples \leq 50 µl. For ssDNA (e.g., single stranded cDNA), add 7 volumes to each volume of sample. e.g., for 50 µl sample add 350 µl Binding Buffer CCB.

Low 260/230 ratio

Possible cause

- Carryover of chaotropic salts

Suggestions

- Give an additional wash step, followed by a dry step at same force and time (step 5).

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® GDNA Cleanup 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® GDNA Cleanup 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® GDNA Cleanup 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.

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