For research purpose only. Not for use in diagnostic procedures for clinical purposes. For IN VITRO USE ONLY.

G-spin™ Genomic DNA Extraction Kit (For Bacteria]

Cat. No. **17121** 50 columns

DESCRIPTION

G-spin™ Genomic DNA Extraction Kit are designed for rapid isolation of genomic DNA from a variety of sample sources including fresh or frozen animal cells/tissues (for Cell/Tissue) and Gram-negative & Gram-positive bacteria (for Bacteria), yeast (for Yeast), or bloods (for Blood). The purified DNA is free of contaminants and impurities and is ideal for all PCR, Southern blotting, RAPD, and sequencing applications.
G-spin™ kit uses advanced silica-gel membrane technology for rapid and efficient purification of genomic DNA without organic extraction or ethanol precipitation. Furthermore, G-spin™ buffer system is optimized to allow rapid and simple cell lysis followed by selective binding of DNA to the column. G-spin™ procedure is very simple, so you can purify DNA from a variety of target source within 20-40min.

STORAGE

- Pre-buffer : store at 4℃.
- G-buffer : store at 4℃. G-buffer has effective efficiency for two months at RT.
- Binding buffer : store at 4℃.
- Washing buffer A, B : store at RT .
- Rnase A Soln, Proteinase K Soln, Lysozyme Soln. : store at -20℃.
- The other components : store at RT

KIT CONTENTS

- Pre-buffer (For Gram-Positive Bacteria) 3ml
  : Before use, add 15㎕ of RNase A stock solution. After add RNaseA, Pre-buffer should then be stored at 2-8?C.
- G-buffer (for positive and negative bacteria) 20ml
  : Before use, add 210㎕ of RNase A stock solution and add 40㎕ of Proteinase K stock solution. After add Rnase A and Proteinase K, G-buffer should then be stored at 2-8℃.
- Binding Buffer 15ml
  : Genomic DNA binding solution. After receiving, Binding Buffer should then be store at 4℃.
- Washing buffer A 9ml
  : Before use, add 21ml of absolute EtOH.
- Washing buffer B 10ml
  : Before use, add 40ml of absolute EtOH.
- Elution buffer 50ml
- Columns 50 columns
  : Columns containing sillica-membrane
- Collection tubes 50 tubes
  : Polypropylene tube for 2ml.
- Lysozyme stock solution (100mg/ml) 200?l
- RNase A stock solution (20mg/ml) 300?l
- Protainase K stock solution (20mg/ml) 40?l

HOMOGENIZATION TECHNIQUES

1. *For bacteria* : Harvest cells in a microcentrifuge tube by centrifuging for 1min at 13,000rpm. Collected cell are resuspended by vortex or repetitive tapping.

PROTOCOL (For Gram-Negative Bacteria)

1. Harvest 1-2ml of cells (OD_{600} : 0.8-1.0) by centrifuging at 13,000rpm for 1min. Remove supernatant.
   Note : After centrifugation, remove the supernatant and completely resuspend by vortex or tapping. Do not to overload the sample.
2. Add 300㎕ of G-buffer solution, and invert-mix well.
3. Incubate at 65℃ for 15min.
   Note : To help lysis cells, invert mix the tube every 5min during the incubation.
4. Add 250㎕ of Binding buffer, and completely mix well by pipetting (at least 10 times) or gently vortexing.
   Note : This step conduces to pass efficiently cell lysates through a column and increase gDNA binding onto column resins and important for efficient deproteinization.
5. Cell lysates loading on column and centrifuge at 13,000rpm for 1min.
   Note : The maximum volume of the column reservoirs 800㎕. For sample volumes of more then 800㎕, sample load and spin again.
6. To wash, add 500㎕ of Washing buffer A to column and centrifuge for 1min at 13,000rpm.
7. Remove solution. Add 500㎕ of Washing buffer B to column and centrifuge for 1min at 13,000rpm.
8. Remove solution and centrifuge for 1min at 13,000 rpm.
9. Place the Gspin™ column in a clean 1.5ml microcentrifuge tube (not provided), and add 50-200㎕ of Elution buffer directly onto the membrane.
10. Incubate at RT for 1min, and then centrifuge for 1min at 13,000rpm.

PROTOCOL (For Gram-Positive Bacteria)

1. Harvest 1-2ml of cells (OD_{600} : 0.8-1.0) by centrifuging at 13,000rpm for 1min. Remove supernatant.
   Note : After centrifugation, remove the supernatant and completely resuspend by vortex or tapping. Do not to overload the sample.
2. Add 50㎕ of Pre-buffer and 3㎕ of Lysozyme solution, mix well.
3. Incubate at 37℃ for at least 15min.
   Note : To help lyse cells, invert mix the tube every 5min during the incubation.
5. Add 250㎕ of G-buffer solution, and invert-mix well.
6. Continue with the "PROTOCOL (For Gram-negative Bacteria)" from step 3.

www.intronbio.com
info@intronbio.com

iNtRON BIOTECHNOLOGY