Genomic DNA Isolation Kit

(Catalog #K281-50; Store at –20˚C)

I. Introduction:

The Genomic DNA Isolation Kit provides a simple and convenient procedure for rapid isolation of genomic DNA from mammalian cells and tissue samples with high yield and purity. The novel method requires less than 90 minutes to prepare highly pure genomic DNA. The extracted genomic DNA is free from protein and RNA, and suitable for a variety of applications such as PCR, DNA hybridization, enzyme manipulation, cloning, Southern blot, and array-based experiments.

II. Kit Contents:

Component	K281-50 50 assays	Cap Color	Part Number
Cell Lysis Buffer	2 x 1.8 ml	Purple	K281-50-1
Enzyme Mix (lyophilized)	1 vial	Red	K281-50-2
TE Buffer	1.5 ml	Green	K281-50-3

III. General Consideration and Reagent Preparation:

- Read the entire protocol before beginning the procedure.
- After opening the kit, store Enzyme Mix at –70˚C. Store buffers at 4˚C.
- Add 275 µl of TE buffer to Enzyme Mix, mix well, aliquot and refreeze immediately at –70˚C. Stable for up to 3 months at –70˚C.
- Be sure to keep all buffers on ice at all times during the experiment.
- The protocol is designed for using with 1 - 2 x 10^6 cells, and generally produces 5 - 20 µg genomic DNA. If larger amount of DNA is desired, scale up the volumes proportionally.

IV. Genomic DNA Isolation Protocol:

1. Collect cells (1 - 2 x 10^6) by centrifugation at 600 x g for 5 min at 4˚C.
   Note: For tissue samples, ground freshly excised tissue in liquid nitrogen. Weigh ~ 5 mg grounded fine tissue powder in a microcentrifuge tube.
2. Add 35 µl of Cell Lysis Buffer. Mix and keep on ice for 1 min. Vortex for 5 sec.
3. Centrifuge in a microcentrifuge tube at top speed for 3 min. Remove supernatant. The pellet is the isolated nuclei.
4. Resuspend the pellet in 40 µl Cell Lysis Buffer.
5. Add 5 µl of Enzyme Mix, pipet several times to mix.
6. Incubate at 50˚C water bath for 1 hr or until the solution becomes clear.
   Notes:
   a. You may extract the sample using 50 µl of Phenol/Chloroform to remove insoluble materials before doing ethanol precipitation (optional).
   b. If isolating DNA for DNA damage quantitation, incubate at 37˚C for 1-2 hrs after adding enzyme mix and extract sample using 50 µl of Phenol/Chloroform.
7. Add 100 µl absolute ethanol, mix and keep at –20˚C for 10 minutes.
8. Centrifuge in a microcentrifuge at top speed for 5 min at room temperature.
9. Remove the supernatant.
10. Wash the DNA pellet 2 times with 1 ml of 70% ethanol. Remove the trace amount ethanol using pipet tip. Air dry for 5 min. (Note: Do not completely dry the DNA. It would be difficult to dissolve if it is completely dried.)
11. Resuspend the DNA in 20 µl TE Buffer or water, store the extracted DNA at –20˚C for future use.

RELATED PRODUCTS:

- Mitochondria/Cytosol Fractionation Kit
- Nuclear/Cytosol Fractionation Kit
- Cytosol/Particulate Rapid Separation Kit
- Membrane Protein Separation Kit
- Mammalian Cell Extraction Kit
- Mitochondrial DNA Isolation Kit
- Link-FAST™ 5 Minutes DNA Ligation Kit
- Gel-FAST™ 20 Minutes Gel Staining/Destaining Kit
- Taq DNA Polymerase
- T4 DNA Ligase
- Apoptosis Detection Kits and Reagents
- Caspase Activity Assay Kits and Active Enzymes
- Mitochondrial Apoptosis Detection Kits and Reagents
- Annexin V Apoptosis Detection Kits and Bulk Reagents
- Beta-Secretase Activity & Drug Screening Kits
- Senescence Detection Kit
- HDAC Activity & Drug Screening Kits
- Akt and JNK Activity Assay Kits & Reagents
- Cell Proliferation & Cytotoxicity Assay Kits
- CETP Activity Assay Kit
- Calpain Activity Assay Kit & Active Enzyme
- Nitric Oxide Detection Kits
- Glutathione Detection Kit
- Quality Antibodies for Apoptosis & Cell Signaling Molecules
- Growth Factors & Cytokines
- Adipocyte secreted Proteins (Adiponectin, Resistin, Leptin, etc.)
- Protease Inhibitor Cocktail

FOR RESEARCH USE ONLY! Not to be used on humans.

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