

Cell & Tissue DNA Extraction Kit

Catalog Number: EXNA075

Size: 50 Tests

For Research Use Only. Not Intended for Diagnostic or Therapeutic Use.



info@exreprotein.com

Product Description

The Cell & Tissue DNA Extraction Kit uses a centrifugal adsorption column that can specifically bind DNA and a unique buffer system for rapid extraction of genomic DNA from animal tissues and cells. The optimized Binding Buffer and Proteinase K rapidly lyses cells and inactivates intracellular nucleases, and genomic DNA is selectively adsorbed on the silicon matrix membrane in the centrifuge column. A series of rapid rinse-centrifugation steps remove cell metabolites, proteins, and other impurities. The purified genomic DNA is compatible with Southern-blot and enzyme digestion reactions.

The kit is efficient and simple-to-use. Single samples generate highly pure genomic DNA in under 30 minutes.

Limitations

- For Research Use Only. Not Intended for Diagnostic or Therapeutic Use.

Precautions

- Always wear appropriate protective clothing and follow safe laboratory procedures.
- Minimize the length of UV exposure when excising the band from the gel to avoid damage to DNA.

Materials Provided & Storage

Shipped at ambient temperature. Proteinase K shipped on blue ice.

Store the kit at 2-30°C for up to 24 months. Proteinase K in glycerol buffer, store at 2-8°C for 12 months or -15 to -25°C for 24 months.

Component	Size	Quantity
Balanced Buffer	5 mL	1 Bottle
Lysate Buffer	11 mL	1 Bottle
Binding Buffer	11 mL	1 Bottle
Inhibitor Removal Solution	25 mL	1 Bottle
Wash Buffer	15 mL	1 Bottle
Elution Buffer	15 mL	1 Bottle
Proteinase K	1 mL	1 Bottle
Adsorption Column and Collection Tube	50 Pcs	1 Bag

Additional Materials Required, Not Supplied

1X PBS	RNase A
Isopropanol	1.5 mL centrifuge tubes
Ethanol (96-100%)	

Preparation of Reagents

Wash Buffer: Add 60 mL ethanol (96-100%) to the Wash Buffer before use.

Protocol

Column equilibration: The adsorption column should be pretreated with Balanced Buffer before the experiment to improve the nucleic acid binding ability. Place the Adsorption Column into the Collection Tube, add 100 μ L of Balanced Buffer to each tube pairing, centrifuged at 13,000 $\times$ g for 60 seconds, discard the waste liquid from the Collection Tube, and place the Adsorption Column back into the Collection Tube for use later.

Cultured Cell Preparation:

1. Add 10^6 - 10^7 suspended cells to 1.5 mL centrifuge tube (do not exceed 10^7 cells). For adherent cells, treat with Trypsin.
2. Centrifuge at 13,000 $\times$ g for 60 seconds to remove the supernatant.
3. Resuspend cells in 200 μ L of 1X PBS, centrifuge at 13,000 $\times$ g for 60 seconds, discard the supernatant, and resuspend the cells in 180 μ L of 1X PBS.
4. Add 20 μ L of Proteinase K solution, mix thoroughly, then add 200 μ L Binding Buffer, vortex thoroughly, and place at 70°C for 10 minutes. **Optional:** if RNA removal is required, add 20 μ L RNase A (25 mg/mL), shake to mix, and leave at room temperature for 5-10 minutes after the completion of step 4.
5. Proceed to Extraction Protocol.

Animal Tissue Preparation:

1. Add 180 μ L of Lysate Buffer to fresh or thawed tissue ($\leq$ 25 mg), homogenize on ice.
2. Add 20 μ L of Proteinase K solution and vortex thoroughly.
3. Place the lysates in a water bath at 56°C for 1-3 hours or until tissue digestion is complete, shaking gently several times to assist lysis. **Optional:** if RNA removal is required, add 20 μ L RNase A (25 mg/mL), shake to mix, and leave at room temperature for 5-10 minutes after the completion of step 3.
4. Add 200 μ L Binding Buffer, vortex thoroughly, and place at 70°C for 10 minutes.
5. Proceed to Extraction Protocol.

Extraction Protocol:

1. Allow sample to cool to room temperature. Add 100 μ L isopropanol, immediately vortex to mix thoroughly.
2. Place the Adsorption Column into the Collection Tube. Add the mixture (including the flocculent precipitate) to the Adsorption Column, centrifuge at 13,000 $\times$ g for 60 seconds, and discard the waste liquid.
3. Add 500 μ L of Inhibitor Removal Solution, centrifuge at 12,000 $\times$ g for 60 seconds, and discard the waste liquid.
4. Add 600 μ L of Wash Buffer (per Preparation of Reagents), centrifuge at 12,000 $\times$ g for 60 seconds, discard the waste liquid, and repeat this step once.
5. Place the Adsorption Column back into the Collection Tube and centrifuge at 13,000 $\times$ g for 2 minutes to thoroughly remove the Wash Buffer and minimize residual ethanol affecting the downstream reaction.
6. Place the Adsorption Column in a new 1.5 mL centrifuge tube, add 100 μ L Elution Buffer to the center of the membrane and place it at room temperature for 3-5 minutes.
7. Elute genomic DNA by centrifugation at 12,000 $\times$ g for 60 seconds and store at -20°C. **Note:** the eluted solution can be pipetted back into the Adsorption Column, allowed to stand for 3-5 minutes, and centrifuged at 12000 $\times$ g for 60 seconds to increase the DNA concentration. Preheating the Elution Buffer to 65-70°C in advance may improve results.