

## GENE PREP Application Guide

### DNA Tissue



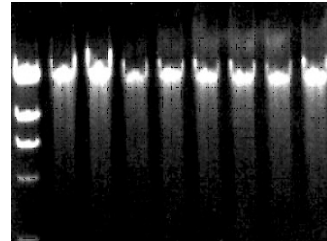
- I. **Purpose:** To extract genomic DNA from mouse tail and other types of animal tissues.
- II. **Chemical Principal:** Proteinase K digestion & organic extraction method.
- IIIA. **Preparation of Tissue Digest (manual ProK digestion for mouse tail):**
  - A. Cut 5 - 10 mm of tail (about 10-20 mg) and place it into AutoGen Tube Unit
  - B. Add 250  $\mu$ l of a buffered Proteinase K solution (250 $\mu$ l of Reagent GPR-1 including 0.8mg/ml Proteinase K) and 250 $\mu$ l of Reagent GPR-2.
  - C. Incubate samples at 55-60°C overnight, and load tube unit onto GENE PREP
- IIIB. **Preparation of Tissue Digest (on-board ProK digestion for mouse tail):**
  - A. Cut 5 - 8 mm of tail (about 10 mg) and place it into AutoGen tube unit
  - B. Load AutoGen Tube Unit onto the GENE PREP
- IV. **Protocol Parameters:**
  - A. **Sample Volume:** 0.5 ml of digest solution.
  - B. **Maximum Number of Samples:** 48 samples per run  
192 samples per run (with an optional Tube Unit Stacker)
  - C. **Processing Time:** 2.1 hours for 48 samples (if digest is prepared manually)  
5.5 hours for 48 samples (if digestion is done on GENE PREP)  
(Includes 40 min drying and re-hydrating time).
  - D. **Yield:** 1 - 2  $\mu$ g of DNA per 1mg of mouse tail (manual digestion).  
0.5 - 1  $\mu$ g of DNA per 1 mg of mouse tail (on-board digestion)
  - E. **Quality:** Typical OD<sub>260/280</sub> values are  $\geq 1.8^*$   
The DNA can be used directly in downstream processes such as fluorescence DNA sequencing, PCR, Southern blotting\* and restriction endonuclease digestions.  
*\* If the tissue is digested on GENE PREP, the OD<sub>260/280</sub> is lower (around 1.7) and the DNA may not be suitable for Southern blotting applications.*

## V. Running the Protocol:

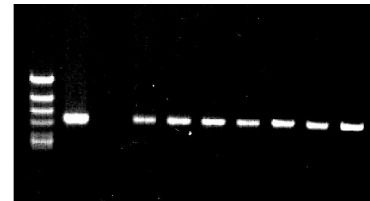
- A. Load Reagents and Samples
- B. Select a Protocol
- C. Enter Number of Samples
- D. Start the Run

## VI. Example of Data:

Genomic DNA obtained from mouse tail  
 Stating Sample: 5-10 mm mouse tail (about 10-20 mg)  
 Digestion: 55°C for 14hrs manually  
 Gel Condition: 5 µl of 100ul resuspended DNA  
 1% agarose gel, 100V x 1hr



PCR of mouse tail DNA  
 Template: 200ng of the above DNA  
 Target Region: ICAM-1 (535bp)  
 PCR Reaction: 35cycles  
 Gel Condition: 10 µl of 25ul PCR reaction  
 1% agarose gel, 100V x 1hr  
 øX174 Hinc II size marker  
 C+/C-: positive and negative controls



## VI. Extraction and Purification Process

Process Site	Purpose	System Process
1. Automated (or Manual)	Digest samples	Dissolve ProK in Reagent TD-M1, and Reagent TD-M2 to sample and incubate at 55-60C.
2. Automated	Precipitate protein	Add Reagent TD-R3, mix and centrifuge. Transfer supernatant to DNA tube.
3. Automated	Precipitate DNA	Add Reagent TD-R4, agitate, centrifuge and discard supernatant.
4. Automated	Wash DNA	Add Reagent TD-R5, mix, centrifuge and discard supernatant. Repeat a few times.
5. Automated	Dry DNA	Transfer DNA tube to agitation rack.
6. Automated	Resuspend DNA	Add Reagent TD-R6, mix and centrifuge.

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