Protocol

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DNA Extraction from Mouse Tail

Utilizing Proteinase K

Introduction

Clean genomic mouse DNA is used for the purposes of genotyping and molecular cloning. Mice are often used as a model organism to study mammals and the tail is a convenient place from which to extract DNA. Sodium Chloride and Sodium Dodecyl Sulfate are used to lyse the cells and Proteinase K serves to prevent degradation of genomic DNA during lysis by digesting nucleases. EDTA also helps to inactivate nucleases by chelating metal ions required correct conformation of the nucleases. Ethanol precipitation is a safe and easy method of isolating genomic DNA.

Materials

- Proteinase K (Proteinase K, GoldBio Catalog # P-480 [CAS 39450-01-6, mw.=28.9 kDa])
- 1M Tris Buffer (See Tris Buffer Stock Solution protocol)
- EDTA Disodium (EDTA Disodium, GoldBio Catalog # E-210 [CAS 6381-92-6, mw. = 372.24 g/mol])
- Sodium Chloride (NaCl) [CAS 7647-14-5, mw. = 58.44 g/mol]
- Sodium Dodecyl Sulfate (SDS) [CAS 151-21-3, mw. = 288.37 g/mol]
- 100% Ethanol
- 70% Ethanol

Method

Stock Solution Preparation

1. Prepare Lysis Buffer stock solution (50 ml).
    a. Add 584 mg of NaCl.
    b. Add 93 mg of EDTA Disodium.
    c. Add 125 mg SDS.
    d. Add 5 ml of 1M Tris Buffer, pH 8.0 into a beaker.
    e. Fill to a final volume of 50 ml with pure H_2O.
    f. Store in aliquots at -20°C.

Sample preparation

1. Add 288 µl of Lysis Buffer to each 1.5 ml microcentrifuge tube.

2. Add 6 µl of thawed Proteinase K Stock Solution, 20 mg/ml (See Proteinase K Stock Solution protocol), to each 1.5 ml tube just before use.

Nuclease Digestion

1. Add a 2 mm length of mouse tail each 1.5 ml tube.
2. Incubate samples at 55°C overnight.

Precipitation

1. Add 1 ml 100% ethanol and mix well.
2. Spin samples with a centrifuge at 16,000 x g for 30 minutes.
3. Remove the supernatant.
4. Wash the pellet with 1 ml of 70% ethanol.
5. Spin the samples with a centrifuge at 16,000 x g for 20 minutes.
6. Remove the supernatant and immediately perform step 12.

Storage

1. Dissolve the DNA pellets in 300 µl of 1X TE buffer (See 10X TE Buffer Stock Solution protocol).
2. Place the samples on heat with the lids open at 55°C for ~2 hours or until all ethanol is evaporated.
3. Store DNA samples at 4°C or -20°C and avoid freeze-thaw cycles.

Tips

- **WARNING: ethanol is flammable. Do not use near open flame.**

Associated Products

- Proteinase K (GoldBio Catalog # P-480)
- EDTA Disodium, dehydrate (GoldBio Catalog # E-210)
- Tris (Tris Base) (GoldBio Catalog # T-400)
- Tris HCl (GoldBio Catalog # T-095)

References

Wang, Z., & Storm, D. R. (2006). Extraction of DNA from mouse tails. *BioTechniques*, *41*(4), 410-412.