

PUREGENE® DNA Purification Kit

DNA Purification From 5-10 mg Mouse Tail Tissue

Expected Yield Range 10-75 µg DNA

Cell Lysis

1. Chill a 1.5 ml tube containing 300 µl **Cell Lysis Solution** on ice. Please note that the solution will turn cloudy.
2. Place 5 mm (5-10 mg) fresh or frozen mouse tail tissue (minced if possible) into the chilled **Cell Lysis Solution**.
3. Add 1.5 µl **Proteinase K Solution** (20 mg/ml) to the sample and mix by inverting 25 times. Incubate at 55°C overnight or until tissue has dissolved. If possible, invert tube periodically during the incubation.

RNase Treatment (optional)

1. Add 1.5 µl **RNase A Solution** to the cell lysate.
2. Mix sample by inverting the tube 25 times and incubate at 37°C for 15-60 minutes.

Protein Precipitation

1. Cool sample to room temperature.
2. Add 100 µl **Protein Precipitation Solution** to the cell lysate.
3. Vortex at high speed for 20 seconds to mix the **Protein Precipitation Solution** uniformly with the cell lysate.
4. Centrifuge at 13,000-16,000 x g for 3 minutes. The precipitated proteins will form a tight pellet. If the protein pellet is not visible, repeat step 3 followed by incubation on ice for 5 minutes, then repeat Step 4.

DNA Precipitation

1. Pour the supernatant containing the DNA (leaving behind the precipitated protein pellet) into a clean 1.5 ml microfuge tube containing 300 µl **100% Isopropanol** (2-propanol).
2. Mix the sample by inverting gently 50 times.
3. Centrifuge at 13,000-16,000 x g for 1 minute; the DNA will be visible as a small white pellet.
4. Pour off the supernatant and drain tube on clean absorbent paper. Add 300 µl **70% Ethanol** and invert tube several times to wash the DNA pellet.
5. Centrifuge at 13,000-16,000 x g for 1 minute. Carefully pour off the ethanol. *Pellet may be loose so pour slowly and watch pellet.*
6. Invert and drain the tube on clean absorbent paper and allow to air dry for 10-15 minutes.

DNA Hydration

1. Add 50 µl **DNA Hydration Solution** (50 µl will give a concentration of 500 µg/ml if the total yield is 25 µg DNA).
2. Rehydrate DNA by incubating sample 1 hour at 65°C and/or overnight at room temperature. If possible, tap tube periodically to aid in dispersing the DNA.
3. Store DNA at 4°C. For long-term storage, store at -20°C or -80°C.