TaqGen®

Product Manual

Taqgen® Tissue Genomic DNA Purification 50 preps
Cat no: MSA950

The Taqgen® Tissue Genomic DNA Purification Kit is suitable for DNA Extraction from Food, Feed, GMO Crops, Meat and Meat products, Human or animal tissue, Cell Culture, Dried Blood Spot.

For research use only.

Not for use in diagnostic procedures.

Description

The TaqGen® Tissue Genomic DNA Purification, a spin column based kit, provide an easy and rapid process for the isolation of the genomic DNA. The purified DNA can be used directly in a variety of downstream applications, including PCR, RealTime PCR, southern blotting and restriction enzyme digestion. The kit can process up to 200mg animal tissue, mouse tail, paraffin-embedded tissue, 5 x 10^6 cultured cells can be readily processed. This kit allows for the single or multiple simultaneous processing of samples.

Product Components and Storage Conditions

Product	Storage condition	Total	Vol/Reaction For 20mg tissue	Vol/Reaction For 200mg tissue
Proteinase K	2°–8°C	1.2mL	20 µL	20 µL
Genomic Digestion Buffer (GDB)	RT	30 ml	280 µL	550 µL
Lysis Buffer	RT	25 ml	200 µL	300 µL
Wash Buffer WB1	RT	10 ml conc	500 µL	500 µL
Wash Buffer WB2	RT	10 ml conc	500 µL	500 µL l
Elution Buffer	RT	20ml	50-100 µL	100-200 µL
Spin Columns	RT	50 pc	1	1

Reconstitution of buffers/reagents:

Buffer/Enzyme name	Procedure
Proteinase K Lyophilized (20mg)	Add 1.2 mL Proteinase buffer
Wash Buffer WB1 (10 ml conc)	Add 30 ml 100% Ethanol
Wash Buffer WB2 (10 ml conc)	Add 30 ml 100% Ethanol

Storage Conditions: Store the Taqgen® Tissue Genomic DNA Purification Kit at 15°–30°C except the proteinase K after reconstitute (2°–8°C).

Safety Information: The Taqgen® Tissue Genomic DNA Purification kit contains guanidine hydrochloride and proteinase K, should be considered harmful and irritants.

Caution: Handle buffer with care. Bleach reacts with guanidine hydrochoride and should not be added to any sample waste.

Before You Begin

Some tissues will require more vigorous disruption techniques than a disposable pestle will allow. For these tissues, a homogenizer is strongly recommended. Tissue that is not homogeneously disrupted may result in poor purification performance. Disrupting tissues with bead beaters or wand homogenizers will cause DNA shearing prior to purification.

Standard protocol for human or animal tissue (200 mg)

1. **Homogenize sample**
   a. Homegenize up to 200mg human or animal tissue and transfer in a 1.5 mL microcentrifuge tube.
2. **Lyse cells**
   a. Add **550µL Genomic Digestion Buffer (GDB)** and **20µL Proteinase K** Solution. Vortex to mix thoroughly.
   b. Incubate at 65°C for 30 min in a water bath. Afterwards, centrifuge the mixture for 5 min (> 10,000 x g) to pellet contaminants and cell debris.
3. **Adjust DNA binding conditions**
   a. Transfer 300µL clear supernatant from step 2 into a 1.5 ml microcentrifuge tube. Do not disturb or transfer any of the insoluble pellet.
   b. Add **200µL Lysis Buffer**. Vortex to mix thoroughly
      Note: A wispy precipitate may form upon the addition of Lysis Buffer. This does not interfere with DNA recovery.
   c. Incubate at 56°C for 10 minutes.
   d. Add 200µL of 100% ethanol.
4. **Bind DNA**
   a. For each sample, place one **SpinColumn** into a Collection Tube. Apply the sample to the column. Centrifuge for **1 min at 8000 rpm (6000 x g)**.Discard the filtrate and reuse the collection tube.
      Note: Gently transfer the lysate/binding buffer mix (~700 µl) to a gDNA Purification Column pre-inserted into a collection tube, without touching the upper column area. Avoid touching the upper have formed during lysis. Any material that touches the upper area of the column, including any foam, may lead to salt contamination in the eluate
5. **Wash and dry silica membrane**
   a. Add **500µL Wash Buffer WB1**. Close the cap and invert a few times, so that the wash buffer reaches the cap. Centrifuge for **1 min at 8000 rpm (6000 x g)**. Discard the filtrate.
   b. Add **500µL Wash Buffer WB2**. Centrifuge **1 min for 8000 rpm (6000 x g)**. Discard the filtrate.
   c. Centrifuge for **2 min at 14,000 rpm (20,000 x g).**
      Note: Residual ethanol is removed during this step.
6. **Elute DNA**
   a. Place the Spin Column into a 1.5 mL microcentrifuge. Add **100µL Elution Buffer**. Incubate at room temperature for 1 min. Centrifuge **1 min at 8000 rpm (6000 x g).**

Protocol for human or animal tissue (20mg)

1. **Homogenize sample**
   a. Homegenize up to 20mg human or animal tissue and transfer in a 1.5 mL microcentrifuge tube.
2. **Lyse cells**
   a. Add **280µL Genomic Digestion Buffer (GDB)** and **20µL Proteinase K** Solution. Vortex to mix thoroughly.
   b. Incubate at 65°C for 30 min in a water bath. Afterwards, centrifuge the mixture for 5 min (> 10,000 x g) to pellet contaminants and cell debris.
3. **Adjust DNA binding conditions**
   a. Transfer 200µL clear supernatant from step 3 into a 1.5 ml microcentrifuge tube. Do not disturb or transfer any of the insoluble pellet.
   e. Add 200µL Lysis Buffer. Vortex to mix thoroughly. Incubate at 56°C for 10 minutes.

      Note: Adjust the volume of Lysis Buffer based on the amount of starting material.

      Example: If you transfer 200µL of clear Supernatant then add 200 µL lysis Buffer and the add 200µL of 100% ethanol. Proceed with step 4 of standard protocol.

Protocol for cultured cells

1. **Sample Preparation**
   a. Frozen cell samples should be thawed before starting this protocol. Pellet the cells by centrifugation. Wash the cells with cold PBS (4°C). Resuspend cells in 200µL PBS. Proceed to Step 2 of (**Protocol for human or animal tissue-20mg).**
   b. For cells grown in suspension, pellet 5 x 10^6 by spinning at 1,200 x *g* in a centrifuge tube. Aspirate and discard the supernatant, and wash the cells once with cold PBS (4°C). Resuspend cells in 200µL PBS. Proceed to Step 2 of (**Protocol for human or animal tissue-20mg).**