Cellular DNA Extraction Kit (Cat. TC-02D)

Description:

- This kit is designed for rapidly isolating the highest yields and quality of DNA from cells/tissue samples, offering a simple, fast, environmental-friendly protocol for DNA extractions without using vacuum filtration and toxic organic solvents such as Trizol, phenol or chloroform. This kit is specially designed for preparing the pure DNA from small amounts of sample material for RT-PCR amplification, qPCR, southern blot and other DNA assays. The kit is compatible with cells/tissues from LCM samples.
- **Key features:**
  1. Obtain the highest yield and integrity of DNA within 1 hour.
  2. No filter column or vacuum filtration is required, able to avoid the loss of DNA during extracting.
  3. Suitable for extracting the pure DNA from the small/tiny tissues or LCM samples.

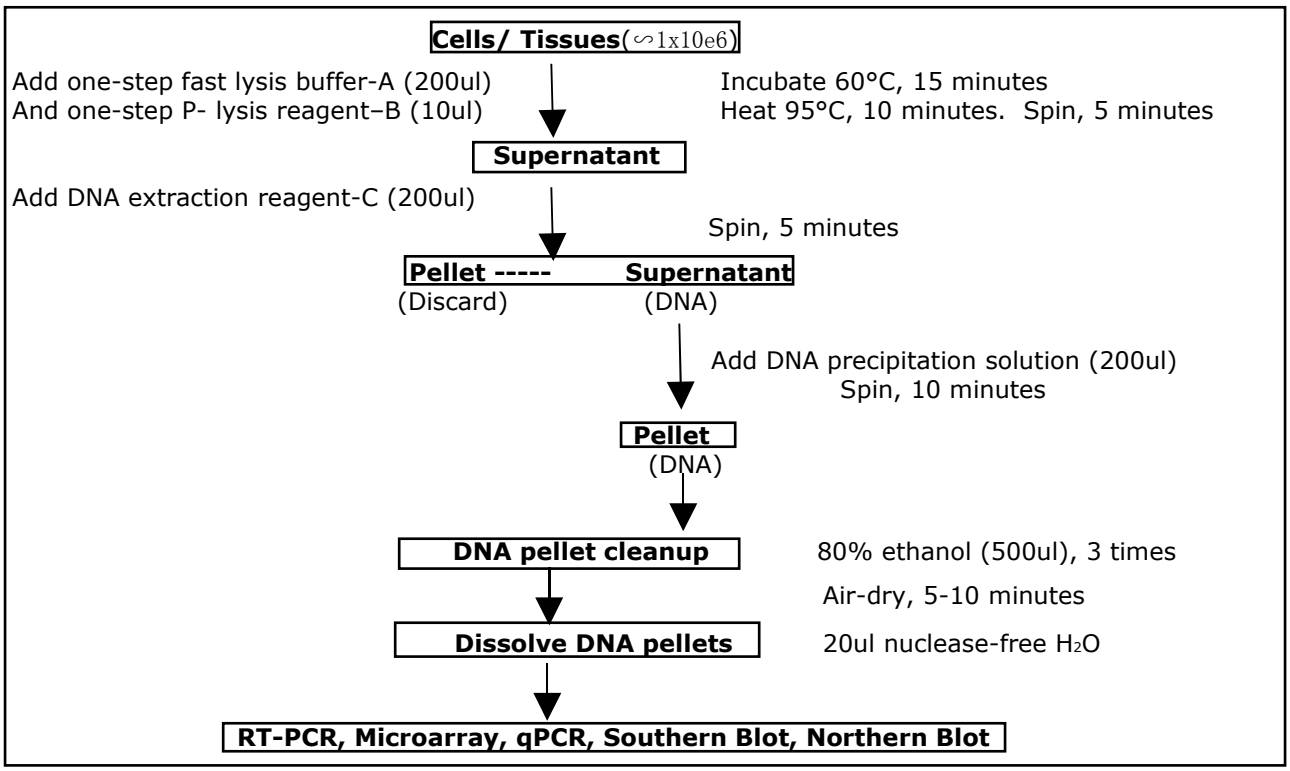
Kit components and protocol: (keep all samples and buffers on ice during proceeding)

Components:	Quantity (100 extracts)	Storage
One-step Fast Lysis Buffer-A	20.0 mL	-20°C
One-step P-Lysis Reagent-B	1.0 mL	-20°C
DNA Precipitation Solution	20.0 mL	-20°C
DNA Extraction Reagent -C	20.0 mL	-20°C

DNA Extraction Protocol:

1. Harvest cells (∽1x10e6) as usual in a 1.5ml microcentrifuge tube, and centrifuge at 1000 xg (or 3000rpm) for 5 minutes at 4°C. Carefully remove all the supernatant and save pellet on ice for DNA extraction.
2. Add **200ul one-step fast lysis buffer-A** and **10ul one-step P-Lysis Reagent-B** to the pellet and incubate on 60°C for 15 minutes. Vortex vigorously at highest speed for 20 seconds every 5 minutes.
3. Heat the samples at 95°C for 10 minutes. Centrifuge at highest speed (∽13,000 xg) for 5 minutes at 4°C. Discard the pellet and transfer the supernatant into a new 1.5ml tube.
4. Add **200ul DNA Extraction reagent-C** into the supernatant, mix thoroughly by pipette up and down several times and centrifuge at 3000xg (∽5,000 rpm) for 5 minute at 4°C. Discard the pellet and transfer the supernatant into a new 1.5ml tube.
5. Add **200ul DNA Precipitation Solution** into the supernatant, mix thoroughly by pipette up and down several times and centrifuge at highest speed (∽13,000 xg) for 10 minute at 4°C. Carefully remove all the supernatant and save the pellet. The pellet is the DNA.
6. Simply rinse the DNA pellets with 500ul ice cold 80% ethanol for 3 times without resuspending the DNA pellet. Aspirate the liquids and Air-dry the pellets for 5-10 minutes.
7. Dissolve the DNA pellets in 20-50ul nuclease-free H_2O or TE. Centrifuge at highest speed (∽13,000 xg) for 2 minute at 4°C prior to measure the DNA concentration with 260/280nm. Store all the DNA extracts at -20°C.
8. Pipette 1-4ul DNA solution into a 25ul PCR mastermix and run PCR/Real-Time PCR at thermal cyclers.

Flow Chart of cellular DNA Extraction: (an innovative DNA precipitation technology)



Precautions and Disclaimer:

This product and procedure described are intended for R&D use only. Purchase of this product does not convey a license to perform any patented process.

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