FFPE Tissue DNA Extraction Kit (Cat. VFFPE-02 D)

Description:

- This kit is designed for extracting DNA from formalin-fixed, paraffin-embedded (FFPE) tissue samples (fresh/frozen tissues, glass slides and blocks) using a simple, rapid, environmental-friendly process without using vacuum filtration and toxic organic solvents such as xylene, phenol or chloroform, prepared for cloning, PCR, qPCR and other DNA assays.
- Key features:
- 1. Obtain highest yield and purity of total nucleic acid within 1.5 hour.
- 2. No filter column or toxic organic solvents required.

Kit contains:

Optimization of zmtech FFPE tissue fast lysis buffers (A&B), including the tissue digestion reagents and a set of complete proteinase enzymes, able to rapidly lysate FFPE tissues and decrease the formalin-induced cross-linking in the sample. Zmtech DNA extraction reagent C, including the DNA precipitation reagents, DNase I inhibitors and PCR reaction enhancers, able to DNA clean-up and enhance PCR reactions.

Kit storage:

Component	Quantity (100 extracts)	Storage
FFPE Tissue Lysis Buffer A	20.0 mL	-20°C
FFPE Tissue Lysis Buffer B	2.0 mL	-20°C Zmtech
DNA extraction reagent C	20.0 mL	-20°C

Additional Reagents required but not provided: Absolute ethanol and 75% ethanol

Protocol:

- 1. Prepare the FFPE sections approximately three or four 20um thick (or 10mg-50mg fresh/frozen paraformaldehyde fixed tissues) in a 1.5 ml clean microcentrifuge tube.
- 2. Add <u>200ul of Lysis Buffer A and 20ul Lysis buffer B</u> into the sample tube. Briefly mix by vortexing and incubate at <u>60°C for 20 minutes</u>. (Vortex for 5 seconds every 10 minutes)
- 3. Place the tube in a PCR machine (or water bath or block) and incubate at <u>95°C for 10 minutes.</u>
- 4. Immediately centrifuge the heated sample tube at maximum speed for 3 minutes at 4°C.
- 5. Pipette the lysate solution into a new sterile/DNAse-free 1.5ml tube by penetrating the thin paraffin layer at the top and not disturbing the pellets (cell debris) at the bottom.
- Centrifuge the lysate solution at 10,000xg for 5 minutes at 4°C. Transfer the supernatant into a new clear 1.5ml tube without disturbing the pellets (the remained cell debris, insoluble tissue fragments, RNA and polysaccharides) at the bottom.
- Add <u>200ul DNA extraction reagent C</u>, mix thoroughly by pipetting up and down. (This mixture may be used for running PCR reactions: Pipette 5-10 ul lysates into a 25ul PCR Master Mixture and run PCR/Real-Time gPCR at thermal cyclers.)
- 8. Add 100ul absolute Ethanol and place the tube on ice for 10 minutes.
- 9. Centrifuge at 14,000 xg for 10 minutes at 4°C. Carefully remove the supernatant, as the DNA pellet might translucent at the bottom of the tube.
- 10. Slowly add <u>500ul 75% ethanol</u> into tube without resuspending the DNA pellet.