TANBead® Nucleic Acid Extraction Kit

OptiPure FFPE DNA Auto Tube

(For use with the SLA-16 / 32 / E13200 series)

RUO

61PS46

(For Research Use Only) V5

1. Intended Use

This product is designed for isolating nucleic acid from various samples, which can be performed by using TANBead® Nucleic Acid Extractor and is intended for research use only.

2. Purpose

TANBead® Nucleic Acid Extraction Kit (61PS46) is used to isolate DNA from formalin-fixed paraffin-embedded (FFPE) tissue samples. This kit is designed to perform the friendly deparaffinization process by using the mineral oil to replace the common hazardous organic solvent, xylene. Typically, using 4 to 10 pieces FFPE samples for extraction, the DNA yield is more than 5 µg (measured by Nanodrop 2000c). The isolated DNA can be applied to molecular analyses, like qPCR, PCR, and next generation sequencing (NGS).

3. The basic principle

The silicon dioxide layer coated on the magnetic beads can adsorb the negatively charged molecules to purify nucleic acids from samples.

4. Specification

Starting Materials	5 µm FFPE section or curl input for 4-10 pieces
Elution Volume	70~100 µL
Typical DNA yield	≧ 5 µg
Typical A260 / A280	1.8 – 2.0

5. Component Supplied with the Kit

96

Auto Tube	8 trays	Auto Tube with reagent buffers
Proteinase K	1.0 mL x 2	Proteinase K
Elution Buffer	1.5 mL x 1	Nuclease-Free Water
Incubation Buffer	35 mL x 1	Tris buffer, surfactants
Mineral Oil	35 mL x 1	Nuclease-Free mineral oil
Base	2	A rack for 8 Auto Tubes
Strip	24	8-channel strip
Protocol	1	Instruction guides for user

6. Auto Tube Content

Well	Buffer	Volume (µL)
1	-	-
2	Washing Buffer 1	500
3	Washing Buffer 2	800
4	Washing Buffer 2	800
5	Magnetic Beads	800
6	Elution Buffer	100

7. Kit Storage and Shelf Life

1) Components under room temperature (15~35°C) can be stored until the expiration date labeled on the box.
2) The proteinase K is transported at room temperature. Upon received, please store proteinase K at 2~8°C.

8. Precautions

1) For research use only.
2) Avoid using expired reagents.
3) When the temperature is below 20°C, place the Auto plates / Auto Tubes in an oven (preheated 42~60°C) 5 to 10 minutes.
4) Avoid vigorous shaking, in order to avoid excessive formation of foam.
5) Carefully remove aluminum foil to avoid splashing.
6) Do not expose the opened reagents or Auto Plates / Auto Tubes to air. The evaporation would lead to pH change, or effect on the extraction effectiveness.
7) Please check the integrity of the Auto Plates / Auto Tubes and remember to insert the spin tips into the appropriate position of the suitable instrument before operating them.
8) Please wear a mask and disposable gloves when handling.
9) Use sterile consumables to avoid nuclease contamination.
10) Reagent solution contains guanidine salt, avoid using bleach containing detergent.
11) Avoid eyes, skin, and clothing contact with reagents. In case of any contact, flush with flowing water.
12) If any serious incident occurs, please report to the manufacturer and the competent authority of the member state in which the user and / or the patient is established.

9. Materials required, Not Supplied

1) TANBead® Nucleic Acid Extraction System
   Model: SLA-16 / 32 / E13200 series (non-sterile)
2) Disposable gloves
3) Scissors, utility knives
4) Micropipette, disposable tips (10 µL / 200 µL / 1000 µL)
5) 1.5 mL microcentrifuge tube
6) IPA: Isopropanol for molecular biology

10. Sample Collection, Storage and Transportation

■ **Sample collection and storage**

The FFPE sample should be stored at 2-25°C. In common, its fixation process, storage time and condition may cause the reduced yield and fragment sizes of nucleic acids.

■ **Sample transportation**

The transportation of FFPE samples should be kept at 2-25°C and follow specific FFPE tissue transportation-related laws.

11. Nucleic Acids Extraction Protocol

■ **Sample preparation**

1) Transfer appropriate FFPE sections into a 1.5 mL microcentrifuge tube.
   Note: 4-8 pieces FFPE sections are suggested to use for common molecular analyses, as for NGS analysis, 8-10 pieces sections may be appropriate.
2) Add **300 µL Mineral oil** and vortex thoroughly, and then incubate at **80°C** for **5 min**.
3) Add **300 µL Incubation Buffer** and **20 µL Proteinase K** and vortex thoroughly, and then centrifuge at **6000 RPM** for **1 min**.
4) The oil-aqueous separated phase will appear and then incubate at **56°C** for **1 – 2.5 hrs**.
   Note: generally, 4-8 pieces sample could be completely lysed in approximate one hour. If few fine residues appeared in lower aqueous phase, please carefully resuspend them by pipet without disturbing the upper layer and incubate for additional 15-30 min. In case of the obvious dissolved residues still exist till 2.5 hours, please keep on step 5.
5) Incubate at **80°C** for **15 min**.
6) Centrifuge at **10000 RPM** for **15 seconds**.
7) Carefully collect the **colorless lower aqueous phase** approximately 200 µL as the sample.

■ **Automatic process**

1) Prepare the Assembled Auto Tubes by inserting Auto Tubes into the Base completely.
2) Carefully remove the aluminum foil from Auto Tubes.
3) Add **200 uL Isopropanol** into well **#1 / #7**, and then transfer the sample **(200 uL colorless lower aqueous phase)** into well **#1 / #7.**
4) Push Assembled Auto Tubes completely to the bottom of the plate rack. Make sure that the chamfer of the plate is at the lower left.
5) Push strips completely to the bottom of strip rack frame, and then close the door panel.
6) Select the program "**LQ-W5-AUTO**". The parameters are given in the following section.
7) Carefully take out Auto Tubes when the program is finished.
8) Use micropipette to transfer the purified nucleic acid from well **#6 / #12** to a clean tube.
9) Discard the used Auto Tubes and strips into the waste recovery can.

12. Program

■ **SLA-16 / 32 series**

Program Name: LQ-W5-AUTO					Model: SLA-16 / 32 series			
Step	Well	Mixing (M)	Collect (S)	Rod	Mixing speed	Volume (μL)	Pause	Vapor (M)
1	5	1	6	On	Medium	800	Off	0
2	1	1	6	On	Medium	800	Off	0
3	2	1	6	On	Medium	800	Off	0
4	3	1	6	On	Medium	800	Off	0
5	4	1	6	On	Medium	800	Off	5
6	6	1	50	On	Medium	150	Off	0
7	5	1	0	Off	Medium	800	Off	0
8	0	0	0	Off	Medium	0	Off	0

■ **SLA-E13200 series**

Program Name: LQ-W5-AUTO						Model: SLA-E13200 series			
Step	Well	Temp (°C)	Mixing (M)	Collect (S)	Rod	Mixing speed	Volume (μL)	Pause	Vapor (M)
1	5	N / A	0.1	6	On	Medium	800	Off	0
2	1	N / A	1	6	On	Medium	800	Off	0
3	2	N / A	1	6	On	Medium	800	Off	0
4	3	N / A	0.5	6	On	Medium	800	Off	0
5	4	N / A	0.5	6	On	Medium	800	Off	5
6	6	N / A	1	50	On	Medium	150	Off	0
7	5	N / A	0.1	0	Off	Medium	800	Off	0
8	0	N / A	0	0	Off	Medium	0	Off	0

13. Reagent performance

■ **Repeatability**

Under repeatability conditions where nucleic acids are extracted with the same reagent kit on the same source samples by the same operator. The coefficient of variation of nucleic acid extraction concentration is less than 5%.

■ **Reproducibility**

A five-day reproducibility test was carried out with the same source samples for 5 consecutive days with the same reagent kit by different operators. The coefficient of variation of nucleic acid extraction concentration is less than 5%.

■ **Detection limit of FFPE DNA samples: ≥ 5 μm**

■ **The stability of extracted DNA**

Storage Conditions	DNA stability
-80°C	Over 90 days
-20°C	28 days
4°C	14 days
25°C	2 days
Freeze-thaw	10 times

14. Explanation of Symbols

	Manufacturer		Consult instructions for use
15°C 35°C	Temperature limit	Σ	Contains sufficient for test
REF	Catalogue number		Caution
LOT	Batch code	NON STERILE	Non-sterile
	Do not re-use		Keep away from sunlight
	Date of manufacture		Use-by date
RUO	For research use only		

Taiwan Advanced Nanotech Inc.
1-4F., No. 8, Ln. 10, Sec. 2, Zhongfu Rd., Zhongli Dist., Taoyuan City 320, Taiwan(R.O.C)
Tel: +886-3-4556599 Revised: 2022-05-17 V5