TANBead® Nucleic Acid Extraction Kit

Plasmid Extraction Auto Plate

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

RUO

6PEA46

(For Research Use Only) V1

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 (6PEA46) is designed for a simple and convenient method of isolating high-quality plasmid DNA from *E. coli* strains including DH10B, DH5α, BL21(DE3), and TOP10. This kit, with SLA-16/32/E13200, utilizes the unique non-alkaline lysis method to simplify the pre-treatment protocol of plasmid extraction. It has no repetitive centrifugation steps, reducing time for manual processing and error rate. The purified plasmid can be directly analyzed (such as Nanodrop, PCR, agarose gel electrophoresis, etc.) and used for downstream experiments.

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	*E. coli* culture (OD_{600} 1 - 5)
Elution Volume	50 - 80 µL
Typical DNA yield	Up to 10 µg
Typical A260/A280	1.7 - 2.0

5. Component Supplied with the Kit

Auto Plate	6	Auto Plate with reagent buffers
Incubation Buffer	35 mL x 1	Tris buffer, surfactants
Elution Buffer	1.5 mL x 1	Nuclease-Free Water
RNase A	100 µL x 1	-
Lysozyme	1	Add 100 µL sterile water before use.
Strip	12	8-channel strip
Protocol	1	Instruction manual

6. Auto Plate Content

Well	Buffer	Volume (µL)
1 / 7	Binding Buffer	600
2 / 8	Washing Buffer 1	800
3 / 9	Washing Buffer 2	800
4 / 10	-	-
5 / 11	Magnetic Beads	800
6 / 12	Elution Buffer	80

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 RNase A and Lysozyme are transported at room temperature. Upon received, please store RNase A and Lysozyme at 2-8°C.
3) Incubation buffer should be stored in 4°C after the addition of Lysozyme and RNase A.

8. Precautions

1) For research use only.
2) Avoid using expired reagents.
3) When the temperature is below 20°C, place the Auto Plates 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 to air. The evaporation would lead to pH change, or effect on the extraction effectiveness.
7) Please check the integrity of the Auto Plates 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: Maelstrom SLA-16/32 & SLA-E13200 (non-sterile)
2) Disposable gloves
3) Micropipette, disposable tips (10 µL/ 200 µL/ 1000 µL)
4) 1.5 mL microcentrifuge tube
5) 15 mL / 50 mL conical tube

10. Sample Collection, Transportation, and Storage

■ **Sample collection**

1) The plasmid recovery is directly affected by host strain, antibiotic selection, culture broth, plasmid copy number, plasmid size, insert toxicity, etc. Handling the transformation and enrichment of *E. coli* properly to ensure maximum plasmid yield.
2) We suggest using LB (Luria Bertani) as the culture medium for plasmid isolation. The best starting biomass of *E. coli* is an OD_{600} of 1–5, which should be incubated overnight at 37°C.
3) We do not recommend using *E. coli* culture during cold storage as material, it may lead to the contamination of genomic DNA.

11. Nucleic Acids Extraction Protocol

Before operating, turn on the warm-up system of TANBead® Nucleic Acid Extractor, if it is equipped with temperature controller, please setting at **60°C.**

1) Add **87.5 µL** Lysozyme and **100 µL** RNase A into Incubation Buffer.
2) Harvest the overnight culture of *E. coli* into a 1.5 mL tube by centrifugation at 6,000 × *g* for 2 min, then carefully remove the supernatant.
3) Add 300 µL incubation buffer into a 1.5 mL tube and resuspended the pellet.
4) Incubate the sample at 65°C for 5 min.
5) Centrifuged at 13,000 × *g* for 3 min.
6) Carefully remove the aluminum foil on the Auto Plates.
7) Transfer 300 µL suspension sample into **well #1 / #7** of Auto Plate (Plate filled with binding buffer).
8) Push auto plate completely to the bottom of plate rack. Make sure that the chamfer of the plate is at the lower left.
9) Push strips completely to the bottom of strip rack frame.
10) Close the door panel.
11) Select the program "**6PE**". The parameters are given in following section.
12) Carefully remove the Auto Plate when the program is finished.
13) Use micropipette to transfer the purified nucleic acids from **well #6 / #12** to a clean tube.
14) Discard used Auto Plate and strips into the waste recycling bin.

12. Program

■ **SLA-16/32 series**

Program Name: 6PE					Model: SLA-16/ 32 series			
Step	Well	Mixing (M)	Collect (S)	Rod	Mixing speed	Volume (μL)	Pause	Vapor (M)
1	5	0	12	ON	Medium	800	Off	0
2	1	3	12	ON	Medium	900	Off	0
3	2	1	12	ON	Medium	800	Off	0
4	3	1	12	ON	Medium	800	Off	0
5	5	1	12	ON	Medium	800	Off	5
6	6	5	60	ON	Fast	100	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:6PE						Model: SLA-E13200 series			
Step	Well	Temp (°C)	Mixing (M)	Collect (S)	Rod	Mixing speed	Volume (μL)	Pause	Vapor (M)
1	5	-	0	12	ON	Medium	800	Off	0
2	1	60	3	12	ON	Medium	900	Off	0
3	2	-	1	12	ON	Medium	800	Off	0
4	3	-	1	12	ON	Medium	800	Off	0
5	5	-	1	12	ON	Medium	800	Off	5
6	6	60	5	60	ON	Fast	100	Off	0
7	5	-	1	0	OFF	Medium	800	Off	0
8	0	-	0	0	OFF	Medium	0	OFF	0

13. Result

Total nucleic acid yield and purity were detected using Nanodrop spectrophotometers: Up to 10 μg DNA per test and the A260/A280 ratio of the nucleic acid is between 1.7 to 2.0.

14. Reagent performance

■ **Host strain**

Several *E. coli* strains including DH10B, DH5α, BL21(DE3), and TOP10 can be used for plasmid isolation by this kit.

■ **Plasmid type**

Common cloning vector plasmids (such as pUC, pBS, pBR and pET) can be isolated by this kit.

■ **Plasmid size**

The plasmid which is less than < 15 kb can be isolated by this kit, isolating larger size of plasmid may decrease the yield of plasmid.

■ **Extraction time and throughput**

Less than 28 min / 16/32 preps on SLA 16/32 series.
Less than 26 min / 32 preps on E13200 series.

15. Explanation of Symbols

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

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