

**OptiPure cfDNA Auto Plate** 

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

(For Research Use Only) V2

### 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 (61CA46) is suitable for extracting cell-free DNA from plasma or serum of human species. Sample need to be pre-treated with proteinase K and SDS before automated nucleic acid extraction process by SLA-16 / 32 / E13200 series. The isolated cfDNA can be directly used for real-time PCR. With highly sensitivity, this reagent kit can be applied for research.

### 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	300 μL plasma / serum
Elution Volume	70~100 μL
Typical DNA yield	1~100 ng per mL plasma / serum
Typical DNA size	100~300 bp

 $\triangle$  DNA yield will vary from 1~100 ng according to sample source and storage method. The average DNA yield is 20 ng/ mL of serum from healthy individuals.

### 5. Component Supplied with the Kit

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Auto Plate	6	Auto Plate with reagent buffers				
Proteinase K	1.0 mL x 2	Proteinase K				
Elution Buffer	1.5 mL x 1	Nuclease-Free Water				
Strip	12	8-channel strip				
Protocol	1	Instruction guide for user				

### 6. Auto Plate Content

Well	Buffer	Volume (µL)
1/7	Lysis Buffer	800
2/8	Washing Buffer 1	1000
3/9	Washing Buffer 2	1000
4 / 10	Washing Buffer 2	1000
5 / 11	Magnetic Beads	1000
6 / 12	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.
- When the temperature is below 20°C, place the Auto Plates / Auto 3) 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 strips 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) 15 mL / 50 mL conical tube
- 7) 20% SDS (Sodium dodecyl sulfate)
- 8) Streck Cell-Free DNA BCT tubes or other Cell-Free DNA collection tube

### 10. Sample Collection, Transportation, and Storage

- Sample collection and storage
- 1) Plasma

- a. Blood samples collected in Streck Cell-Free DNA BCT tubes is recommended. It is stable for up to 14 days.
- b. Centrifuge whole blood at 1600 x g for 10 minutes.
- c. Transfer the upper plasma layer to a new conical tube.
- d. Centrifuge at 16000 x g for 10 minutes.
- Carefully transfer the supernatant to a new conical tube for e. cfDNA isolation.
- 2) Serum
  - a. Blood samples collected in Streck Cell-Free DNA BCT tubes is recommended. It is stable for up to 14 days.
  - b. Let the blood clot for at least 30 minutes in room temperature.
  - c. Centrifuge whole blood at 1600 x g for 10 minutes.
  - d. Transfer the upper serum layer to a new conical tube.
  - e. Centrifuge at 16000 x g for 10 minutes.
  - f. Carefully transfer the supernatant to a new conical tube for cfDNA isolation.
- 3) Specimen storage
  - a. After centrifugation, the plasma / serum can be stored at
    - i. 2~8°C up to 7 days.
    - ii. -20°C for long-term preservation.

### Specimen transportation

Transportation of whole blood, serum specimens should be followed by specific pathogen transportation-related laws. The whole blood sample should be kept between 2~25°C during transportation and within 6 hours for separated serum. Plasma / Serum samples can be transported between 2~8°C or by freezing.

### 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 25°C.

- 1) Add 300 µL plasma / serum with 30 µL 20% SDS and 20 µL PK into microcentrifuge tube
- Mix well and incubate at 60°C for 20 minutes. 2)
- 3) Carefully remove the aluminum foil on the Auto Plates.
- Add total mixture into well #1 / #7 of 96 well plate.
- 5) Push Auto Plates completely to the bottom of the plate the rack. Make sure that the chamfer of the plate is at the lower left.
- 6) Push strips completely to the bottom of strip rack frame.
- 7) Close the door panel.
- 8) Select the program "CFDNA". The parameters are given in following section.
- 9) Carefully remove the Auto Plates when the program is finished.
- 10) Use micropipette to transfer the purified nucleic acids from well #6 / #12 to a clean tube.
- 11) Discard used Auto Plates and strips into the waste recycling bin.



## 12. Program

### SLA-16 / 32 series

Program Name: CFDNA				Model: SLA-16 / 32 series				
Step	Well	Mixing (M)	Collect (S)	Rod	Mixing speed	Volume (µL)	Pause	Vapor (M)
1	5	1	30	On	Medium	1000	Off	0
2	1	10	30	On	Very Low	1200	Off	0
3	3	10	0	Off	Medium	1000	Off	0
4	5	1	0	Off	Fast	1000	Off	0
5	3	0	60	On	Medium	1000	Off	0
6	2	3	0	Off	Medium	1000	Off	0
7	5	1	0	Off	Fast	1000	Off	0
8	2	0	60	On	Medium	1000	Off	0
9	4	5	30	On	Medium	1000	Off	5
10	6	4	60	On	Medium	100	Off	0
11	5	1	0	Off	Medium	1000	Off	0

### SLA-E13200 series

Program Name: CFDNA					Model: SLA-E13200 series				
Step	Well	Temp (°C)	Mixing (M)	Collect (S)	Rod	Mixing speed	Volume (µL)	Pause	Vapor (M)
1	5	45	1	30	On	Medium	1000	Off	0
2	1	45	10	30	On	Very Low	1200	Off	0
3	3	45	10	0	Off	Medium	1000	Off	0
4	5	45	1	0	Off	Fast	1000	Off	0
5	3	45	0	60	On	Medium	1000	Off	0
6	2	45	3	0	Off	Medium	1000	Off	0
7	5	45	1	0	Off	Fast	1000	Off	0
8	2	45	0	60	On	Medium	1000	Off	0
9	4	45	5	30	On	Medium	1000	Off	5
10	6	45	4	60	On	Medium	100	Off	0
11	5	NA	1	0	Off	Medium	1000	Off	0

### 13. Result

Total DNA yield was quantified using Qubit dsDNA HS assay kit: 1~100 ng per mL serum / plasma. The expected peak bp (100~300 bp fragments) and low amount of gDNA contamination were observed by using Qsep 100 capillary gel electrophoresis system.



### 14. 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%.

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### 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	5 times

### 15. Explanation of Symbols

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

