TIANGEN®

TGuide S96 Circulating DNA Maxi Kit

www.tiangen.com/en

This product is for scientific research use only. Do not use in medicine, clinical treatment, food or cosmetics.

TGuide S96 Circulating DNA Maxi Kit

Cat. no. GDP810

Kit Contents

Contents	GDP810 (24 preps)
Lysis Enhancer	10 ml
Buffer GHH	1 plate (3 ml /well)
Buffer GDFP	1 plate (2 ml /well)
Buffer GDFP	1 plate (1 ml /well)
Buffer PWGP	1 plalte (2 ml /well)
MagAttract Suspension ESP2	1 plate (550 μl /well)
Proteinase K	3×1 ml
Buffer TBC	15 ml
S96 Deep Well 24 Plate	1 plate
S96 24-Tip Comb with Plate	1 set
Handbook	1

Storage

This kit can be stored at room temperature (15-30°C) under dry condition for 12 months. If a precipitate has formed in Buffer, please place the buffer at 37°C for 10 min to dissolve the precipitate.

Introduction

The kit adopts magnetic beads with unique separation function and a unique buffer system to separate and purify high-quality circulating nucleic acid from samples such as serum/plasma. The unique embedded magnetic beads have strong affinity for nucleic acid under certain conditions, and when the conditions change, the magnetic beads will release adsorbed nucleic acid, thus achieving the purpose of fast separation and purification of nucleic acid. The whole process is safe and convenient, with high yield of free nucleic acid extraction, high purity and stable and reliable quality, and is suitable for meet various downstream detection experiments and NGS analysis.

Features

1. The kit can be perfectly matched with the 24 flux module of TGuide S96 Automated Nucleic Acid Extractor to process the extraction of circulating nucleic acid from large-volume samples.
2. The products obtained by the kit meet the requirements of downstream detection experiments and NGS analysis.

Notes **Please read these notes before using this kit.**

1. Avoid repeated freezing and thawing of the sample, otherwise the extracted nucleic acid fragments will be smaller and the extraction yield will be reduced.
2. If there is precipitation in Buffer GHH, it can be redissolved in a 37 water bath and shake to mix before use.
3. The single extraction volume of this kit is 2 ml.

Preparation of Carrier RNA solution (self-provided)

Carrier RNA is a kind of nucleic acid capture. When the content of nucleic acid in the sample is low, Carrier RNA can improve the extraction efficiency of circulatin nucleic acid.

Add 310 μl of RNase-Free ddH_2O to a tube filled with 310 μg Carrier RNA freeze-dried powder (TIANGEN, Cat# GRT416-02), completely dissolve Carrier RNA to obtain a solution with a final concentration of 1 μg/μl, and aliquot the solution into RNase-free centrifuge tubes according to experimental conditions and store at -30~-15°C.

Note: When using, take out the corresponding solution according to the extraction times. Avoid repeated freezing and thawing, and the freezing and thawing times should not exceed 3 times.

Protocol

This procedure is suitable for processing 2 ml plasma samples using TGuide S96 Automated Nucleic Acid Extractor.

I. Preparation of reagents

Take out the vacuum package pre-packaged 24-deep well plate from the kit, mix it upside down for several times to resuspend the magnetic beads, remove the vacuum package, gently swing the 24-deep well plate to concentrate the reagent and magnetic beads to the bottom of the 24-deep well plate (or centrifuge at 500 rpm for 1 min pusing a plate centrifuge), carefully tear off the sealing film before use to avoid vibration of the 24-deep deep well plate and prevent liquid spillage.

II. Sample treatment (optional)

1. Solution 1 (for higher nucleic acid yield)

Balance the plasma sample to room temperature, add 100 μl Proteinase K, 2 ml plasma sample and 100 μl Lysis Enhancer in turn into the centrifuge tube **(please add the solution in strict accordance with this order, for direct mixing of Proteinase K and Lysis Enhancer will reduce the reaction activity)**. Mix by inverting upside down for 10 times, and incubate at 60°C for 20 min after mixing. After the incubation, take out the tube and place on ice for 5 min or room temperature for 10 min to cool.

2. Solution 2 (fast extraction)

   Allow the plasma sample to balance to room temperature, and add 2 ml of plasma sample and 100 µl Proteinase K to the deep well plate of Buffer GHH.

III. Reagent and plate distribution

1. Add the above-mentioned processed sample and 1 µl Carrier RNA solution (provided by the customer, please refer to the preparation of Carrier RNA solution) into the deep well plate of Buffer GHH.
2. Transfer 100 µl of Buffer TBC into an empty 24-deep well plate.
3. Place the 24-deep well plate into the corresponding position according to the following table

Plate position	E	F	G	H
Reagent	GDFP 2 ml	GDFP 1 ml	PWGP 2 ml	ESP2 550 µl
Plate position	**A**	**B**	**C**	**D**
Reagent	GHH 3 ml Sample 2 ml Proteinase K 100 µl (Carrier RNA 1 µl)	S96 24-Tip Comb	—	TBC 100 µl

4. Run the program for large volume free nucleic acid extraction (see the following table for the program).

Step	Name	Plate	Mixing volume (μl)	Mixing speed	Mixing time (min)	Precipi-tation time (sec)	Adsor-bing times	Adsor-bing speed (mm/s)	Heating posi-tion	Heating temp-eratu (°C)	Suspe-nsion time (min)	Capture action
1	Capture Magnetic rod comb	B	—	—	—	—	—	—	—	—	—	Capture
2	Magnetic beads adsorbing	H	—	—	—	—	1	1.0	—	—	—	—
3	Lysis and binding	A	5000	Middle-slow	25	60	2	0.8	—	—	—	—
4	Washing 1	E	2000	Medium	2	60	1	1	—	—	—	—
5	Washing 2	F	1000	Medium	2	60	1	1	—	—	—	—
6	Washing 3	G	2000	Medium	2	60	1	1	—	—	—	—
7	Washing 4	H	550	Medium	2	60	1	1	—	—	5	—
8	Elution	D	100	Medium	8	60	2	1	D	56	—	—
9	Discard magnetic beads	H	—	—	—	—	—	—	—	—	—	Release

5. After the automatic extraction procedure is completed, transfer the nucleic acid solution in Buffer TBC plate to the centrifuge tube and store under appropriate conditions.