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Magnetic Circulating DNA Maxi Kit V2

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Magnetic Circulating DNA Maxi Kit V2

Cat. no. GDP720

Kit Contents

Contents	GDP720-01 (2 ml × 50 preps)	GDP720-02 (2 ml × 200 preps)
Lysis Enhancer	10 ml	25 ml
Buffer GHH	2 × 80 ml	4 × 160 ml
Buffer GDF	150 ml	3 × 150 ml
Buffer PWG	40 ml	3 × 40 ml
Buffer TBC	15 ml	30 ml
Proteinase K	5 × 1 ml	2 × 10 ml
MagAttract Suspension E	3 × 1 ml	12 × 1 ml
Handbook	1	1

Optinal Products

Carrier RNA (Catalog No. GRT416-02); Magnetic Frame (Catalog No. OSE-MF-01)

Storage condition

The kit can be stored for 15 months at room temperature (15-30°C) in a dry condition. In case of precipitation in the solution, it can be preheated in a 37°C water bath for 10 min to dissolve the precipitation before use, and its effect will not be affected.

Product introduction

The kit can be used to separate and purify high-quality free DNA from serum, plasma and other samples through the magnetic beads with unique separation function and unique buffer system. The uniquely embedded magnetic beads have strong affinity for nucleic acids under certain conditions. When the conditions change, the magnetic beads release the adsorbed nucleic acids, which can achieve the purpose of rapid separation and purification of nucleic acids. The whole process is safe and convenient, and the extracted free DNA has high yield, high purity, stable and reliable quality. It is especially suitable for automatic extraction in high-throughput workstations.

Features

1. This kit can be used for manual extraction or batch extraction on a variety of high-throughput platforms.
2. The products obtained by this kit can meet all kinds of downstream detection experiments and NGS analysis.
3. This product is applicable to 1-2 ml of serum and plasma samples.

Notes <u>Please be sure to read these notes before using this kit.</u>

1. Repeated freezing and thawing of samples should be avoided, otherwise the extracted nucleic acid segments will be relatively small and the extraction amount will be reduced.
2. If there is precipitation in the Buffer GHH, it can be re-dissolved in a 37°C water bath, and then shaken well before use.

Preparation of Carrier RNA solution

Before using for the first time, dissolve the Carrier RNA (310 μg) in 310 μl RNase-free H_2O, and a stock solution with 1 μg/μl concentration is prepared. Aliquot the solution into appropriate number of tubes and store at (-15~-30°C). Avoid repeated freezing and thawing, and the freezing and thawing times should not be more than 3 times.

Protocol

Please add ethanol (96-100%) to Buffer GDF and Buffer PWG as indicated on the bottle tag.

I. Manual operation steps

1. Sample treatment:

A. Sample treatment scheme I (high yield)

1) Balance the plasma sample to room temperature,choose the suitable tubes according to the volume of sample from the form as below and add Proteinase K, plasma sample and Lysis Enhancer in turn.

Tube Size	Proteinase K (µl) 0.05 x Sample Volume	Sample Volume (µl)	Lysis Enhancer (µl) 0.05 x Sample Volume
5 ml	50	1000	50
	75	1500	75
	100	2000	100
15 ml	150	3000	150
	200	4000	200
	250	5000	250

Note: It is recommended to add the solution strictly in this order, otherwise the activity of Proteinase K directly mixed with Lysis Enhancer will decrease.

2) Convert the tube for 10 times to mix it well. After mixing, incubate it at 60°C for 20 min. After incubation, place it on ice for 5 min or at room temperature for 10 min to cool.

B. Sample treatment scheme II (rapid)

Balance the plasma sample to room temperature, choose the suitable tubes according to the volume of sample from the form as below and add Proteinase K, Buffer GHH and MagAttract Suspension in turn, mix them fully and incubate it for 15 min at room temperature, during that period, shake it every 5 min., mix them upside down, incubate them at room temperature for 15 min, and shake and mix them every 5 min.

2. According to the following table, for samples with different volumes, add corresponding volumes of Buffer GHH, MagAttract Suspension E and 1 µl Carrier RNA (self-provided by the customer), mix by vortex for 30 sec, then incubate at room temperature for 15 min to make the magnetic beads adsorb nucleic acid, during that period, mix by vortex for 30 sec every 5 minutes.

   Note: The MagAttract Suspension E should be mixed by vortex for 1-2 min before use to ensure that the magnetic beads are completely mixed.

Tube Size	Sample Volume (µl)	Proteinase K (µl) 0.05 x Sample Volume	GHH (µl) 1.5 x Sample Volume	MagAttract Suspension E lume (µl)
5 ml	1000	50	1500	30
	1500	75	2250	45
	2000	100	3000	60
15 ml	3000	150	4500	90
	4000	200	6000	120
	5000	250	7500	150

3. Place the tube on a Magnetic Frame for 2 min, and remove the liquid carefully with a pipette when the magnetic beads are fully attached. Then remove the tube from the Magnetic Frame.

4. Add 2 ml of Buffer GDF **(ensure that ethanol has been added before use)**, mix by vortex for 2 min to fully suspend the magnetic beads.

5. Place the centrifuge tube on the Magnetic Frame for 1 min, and carefully remove the liquid with a pipette when the magnetic beads are completely adsorbed.

6. Add 1 ml of Buffer GDF and mix by vortex for 2 min to fully suspend the magnetic beads.

7. Place the centrifuge tube on the Magnetic Frame for 1 min, and carefully remove the liquid with a pipette when the magnetic beads are completely adsorbed.
8. Add 2 ml of Buffer PWG **(ensure that ethanol has been added before use)**, mix by vortex for 2 min to fully suspend the magnetic beads.
9. Place the centrifuge tube on the Magnetic Frame for 1 min, and carefully remove the liquid with a pipette when the magnetic beads are completely adsorbed.
10. Add 500 µl of Buffer PWG, mix by vortex for 2 min to fully suspend the magnetic beads, and centrifuge briefly to remove the droplets on the inner wall of the tube cap.

    Note: All magnetic beads and liquid in step 10 could be transferred to a 1.5 ml centrifuge tube for subsequent operation on a 1.5 ml Magnetic Frame.
11. Place the centrifuge tube on the Magnetic Frame for 1 min, and carefully remove the liquid with a pipette when the magnetic beads are completely adsorbed.
12. Place the centrifuge tube on the Magnetic Frame and dry it at room temperature for 5-10 min.

    Note: The residual ethanol will inhibit the subsequent enzyme reaction, so it is necessary to ensure that the ethanol volatilizes completely when drying. But do not over-dry the beads, or it will be hard to elute the nucleic acid.
13. Add 30-100 µl of Buffer TBC, re-suspend the magnetic beads by pipetting, incubate at 56°C for 5 min, and gently shake every 2 min to fully elute the nucleic acid.
14. Place the centrifuge tube on the Magnetic Frame for 2 min. When the magnetic beads are completely adsorbed, carefully transfer the nucleic acid solution to a new centrifuge tube and store it at -20°C.

    Note: The magnetic adsorption time could be appropriately extended, or the nucleic acid solution could be transferred to a new centrifuge tube by high-speed centrifugation for 2 min after the magnetic absorption, so as to ensure that there is no magnetic bead residue in the solution, to avoid affecting the subsequent experiments.

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