



ZGeneBio Urine Circulating Nucleic Acid Extraction Kit

Cat# ZGCIR03-20 Size : 20Reactions

For concentration and purification of
Circulating DNA, RNA and micro RNA from Urine

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Kit Contents

Urine Circulating Nucleic Acid Extraction Kit	20 reaction
Binding Buffer	210 ml
Nano Beads	700 ul
Proteinase K (20 ug/ml)	7 ml
Wash Buffer I	18 ml
Wash Buffer II	22 ml
Elution Buffer	5 ml

Storage

- I. Free Circulating Nucleic Acid Extraction Kit can be stored at 15–25°C upon arrival and are stable for at least one year after delivery.
- II. The kit contains a ready-to-use proteinase K solution, which is dissolved in a specially formulated storage buffer. The proteinase K is stable at room temperature (15–25°C). To prolong the lifetime of proteinase K, storage at 2–8°C is recommended

Introduction

Raised levels of cell-free circulating DNA in cancer patients have been reported in many tumor types and analysis of circulating DNA can provide a useful marker for earlier cancer detection. Reported circulating DNA has a quite small fragment size which is about 200bp. Based on the high efficiency and specificity between nucleic acid and **ZGeneBio** beads. **ZGeneBio** developed the Free Circulating Nucleic Acid Extraction Kit from Urine.

The human urine have been shown to contain cell-free DNA. For urine DNA detection, several recent studies addressed the need for careful evaluation and standardization of preanalytical processes. Key problems appear, such as possible contamination of cells; the generally low and variable amount of free circulating DNA, making extraction/quantification difficult and time-consuming; poor DNA quality; and the presence of PCR inhibitors. In any

case, automation of DNA extraction, which is a prerequisite for introduction of these diagnostic approaches in clinical laboratories, is difficult to achieve because of the volumes of plasma necessary to get sufficient DNA.

The **ZGeneBio** Free Circulating Nucleic Acid Kit is a simple, rapid method and time-saving process, which can be routinely used in laboratories. The

entire procedure takes less than 1 hour as many as 12 samples can be handled in one cycle. It also enables efficient purification of these free circulating nucleic acids and provides high yields from human urine. The kit could starting sample volumes of up to 15 ml, and flexible elution volumes between 100 µl and 200 µl allow concentration of nucleic acid species that are present in low amounts in the sample material. The kit can also be used for purification and concentration of viral nucleic acids from large sample volumes.

Equipment and Reagents to Be Supplied by User

Shaker or rotor

50 ml centrifuge

Water bath or heating block

Ethanol (96–100%)*

Important Notice:

Preparation of buffers and reagents

Wash buffer I :

Add 6 ml ethanol (96–100%) to wash buffer I

Protocols:

Extraction of Free Circulating Nucleic Acids from 10 ml urine:

1. Prepare **10 ml urine** in 50 ml centrifuge tube, then add **10 ml binding buffer, 300 ul proteinase K and 30 ul nano beads**, invert 5 times, then put on the shaker for 100 rpm 10min.
2. 1500 rpm 5min centrifuge. Then Discard the supernatant.
3. Add **1 ml wash buffer I** to pipette beads complete, transfer to a 1.7 ml tube, 13000 rpm 1 min centrifuge, discard the supernatant.
4. Add **1 ml wash buffer II**, pipette beads complete, 13000 rpm 1 min centrifuge, discard the supernatant.
5. After drawing all of wash buffer II off, 3 min at heat block 60C (keep lid open). **Do not over dry.**
6. Add 150 ul elution buffer vortex 20 sec and spin down then 60C water bath 10 min.
7. 14000 rpm 5 min centrifuge. Keep DNA solution for PCR ready.

Extraction of Free Circulating Nucleic Acids from 15 ml urine:

1. Prepare **15 ml urine** in 50 ml centrifuge tube, then add **15 ml binding buffer, 400 ul proteinase K and 35 ul nano beads**, invert 5 times, then put on the shaker for 100 rpm 10min.
2. 1500 rpm 5min centrifuge. Then Discard the supernatant.
3. Add **1 ml wash buffer I** to pipette beads complete, transfer to a 1.7 ml tube, 13000 rpm 1 min centrifuge, discard the supernatant.
4. Add **1 ml wash buffer II**, pipette beads complete, 13000 rpm 1 min centrifuge, discard the supernatant.
5. After drawing all of wash buffer II off, 3 min at heat block 60C (keep lid open). **Do not over dry**.
6. Add 200 ul elution buffer vortex 20 sec and spin down then 60C water bath 10 min.
7. 14000 rpm 5 min centrifuge. Keep DNA solution for PCR ready.

Ordering Information

ZGCIR03-20/50	Urine Circulating Nucleic Acid Extraction Kit
ZGCIR03BD-20/50	Binding Buffer
ZGCIR03NB-20/50	Nano Beads
ZGPTSK-20	Proteinase K (20 ug/ml)
ZGCIR03WI-20/50	Wash Buffer I
ZGCIR03WII-20/50	Wash Buffer II
ZGCIR03EB-20/50	Elution Buffer

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