#### Introduction

The discovery of circulating cell-free DNA (cfDNA) in peripheral blood has been extensively studied and documented. The detected cfDNA can originate from a tumor, fetus or transplanted organs. More recently, it has been determined that small amounts of cfDNA can pass from the blood through the kidney barrier into urine. The obvious advantage of urine sampling makes urine a truly useful source of fetal and tumor cfDNA, a clear advantage over peripheral blood. The cfDNA isolated from urine sample can be used for various applications, including disease monitoring (diagnosis, prognosis and treatment efficacy) and as a screening tool for hereditary conditions.

# **Urine DNA Storage kit**

**Urine DNA Storage kit** are designed for the rapid preservation of nucleic acids from fresh urine specimens. The Urine Storage Tube is a plastic, closed, evacuated tube that contains **x** ml of Urine Preservative additive. The Urine Preservative is a general purpose reagent that contains urine enzyme inhibitors and a cell preservative in a liquid medium. It prevents the growth of Gram-negative and Gram-positive bacteria and fungi, stabilizes nucleated cells, preventing the release of cellular genomic DNA, and inhibits nuclease mediated degradation of circulating cell-free DNA (cfDNA) in urine samples during storage and transportation for up to 7 days at 6 °C to 37 °C. It also inactivates viruses, allowing the resulting non-infectious samples to be handled and shipped safely.

Urine samples preserved in Urine DNA Storage Tube are compatible with most commercially available DNA and cfDNA isolation kits. Samples stored in the tubes have been used successfully with all of CWbioscience's urine-based DNA and cfDNA kits and reagents.

## Precautions

1. For research use only. Not for use in diagnostic procedures.

2. Do not use tubes after expiration date.

3. Do not use tubes if cloudiness or precipitate visible in reagent of empty tube, and contact CWbioscience Technical Services at 4006-222-360 or technicalservices@cwbioscience.com.

4. Do not dilute or add other components to urine DNA storage tube.

5. Avoid contact with skin and mucous membranes.

6. Product and human specimens should be disposed with proper disposal methods in accordance with federal, state and local regulations.

## Storage and stability

1. When stored at 4 °C to 30 °C, unused Urine DNA Storage Reagent is stable through expiration date.

2. Proper insulation may be required for shipment during extreme temperature conditions.

3. Urine samples with the addition of Urine DNA Storage Reagent are stable for up to 7 days when stored between 6  $^{\circ}$ C to 37  $^{\circ}$ C.

## Instructions for use

1. Wash hands with soap and water prior to sample collection.

2. Twist the cap off the Urine Collection Cup in a counterclockwise direction (Do not reveal the protective cover), and invert the cap on a clean table, taking care not to touch the inside of the cap with your fingers.

- 3. Collect urine in the specimen collection cup up to volumes 30-80 ml (about 60 ml). Note: Collect midstream urine. First flow urine is not recommended for collection.
- 4. Screw the cap tightly back onto the cup and mix by gentle inversion 3 to 5 times.
- 5. Remove the protective cover and reveal the collection hole on the top of the cap.

6. Turn Urine DNA Storage Tube upside down and insert into the collection hole, insure the urine sample is fully sucked into the storage tube (about 10ml for each tube).

Note: It is recommended that the urine specimen is transferred into 2 storage tubes within 2 hours of collection.

7. Remove the storage tube from the collection hole and mix by gentle inversion 8-10 times.

8. After collection, transport and store preserved urine samples within the recommended temperature range (6 °C to 37 °C).

#### cfDNA Extraction

- 1. For isolation of cell-free DNA, centrifuge specimens at room temperature at 4000 rpm for 10 minutes, or follow extraction kit manufacturer's instructions.
- 2. Carefully transfer the supernatant to a new tube using a pipette without disturbing the pellet. Follow cell-free DNA extraction kit manufacturer's instructions. CWhipro Circulating DNA Midi Kit (CW2612) is recommended.

## Advantages

- Easy-to-use, painless DNA sample collection
- Quantitative collection of urine specimen by vacuum negative pressure, no need to transfer samples.
- No need to immediately process samples
- Better stability across transit and different temperatures. cfDNA and gDNA are stable for up to 7 days at 6  $^\circ$ C to 37  $^\circ$ C.
- Ship urine samples at room temperature safely
- Compatible with most DNA/RNA isolation methods
- The liquid sample stabilizing reagent is easy to use with liquid handling robots