

ab156899 DNA Isolation Kit - Urine

Instructions for Use

For quick and efficient isolation of DNA from urine.

<u>View kit datasheet: www.abcam.com/ab156899</u> (use www.abcam.cn/ab156899 for China, or www.abcam.co.jp/ab156899 for Japan)

This product is for research use only and is not intended for diagnostic use.

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1. Background

DNA found in urine is mainly derived from cells shed into the urine from the urinary tract. DNA isolated from urine may be used in many different applications in research (ex: DNA methylation identification) and diagnostics (ex: cancer testing). Using urine DNA would have special advantages for diagnostics: (1) the collection of urine is completely non-invasive; (2) technically, isolation of DNA from urine is easier than from blood.

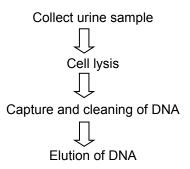
The DNA Isolation Kit - Urine uses a unique procedure and composition to efficiently isolate DNA from urine. The kit also has the following features:

- The fastest procedure available, which can be finished within 20 minutes with consistent isolation conditions.
- High efficiency of DNA isolation from urine.
- Use of non-toxic reagents and no phenol chloroform.

Typical yield of DNA isolated from urine using this kit varies depending on the input sample. It allows isolation of DNA size from 100 bp to 20 kb; DNA quantity from 1 ng to 2 μ g, optimal at between 10 ng and 1 μ g.

ab156899 simply applies our proprietary DNA isolation buffer to urine sediments. After treatment with DNA digestion buffer, the DNA is easily recovered in 8-20 μ L by our specially designed Fast-Spin Columns. DNA is then ready for downstream applications.

2. Assay Summary



3. Materials Supplied

Item	50 tests	100 tests	Storage (Before Preparation)
DNU1 (Suspending Buffer)	16 mL	2 x 16 mL	RT
DNU2 (DNA Digestion Solution)	1.1 mL	2.2 mL	RT
DNU3 (DNA Digestion Powder)	1 vial	2 vials	–20°C
DNU4 (DNA Capture Buffer)	16 mL	2 x 16 mL	RT
DNU5 (DNA Elution Solution)	1 mL	2 mL	RT
F-Spin Column	50	100	RT
F-Collection Tube	50	100	RT

4. Storage and Stability

Upon receipt:

- (1) DNU3 (DNA Digestion Powder) should be stored at -20° C, or stored at 4° C as soon as it is dissolved in DNU2 (DNA Digestion Solution);
- (2) Store all other components at room temperature.

5. Materials Required, Not Supplied

- Waterbath or heat block
- Vortex mixer
- Desktop centrifuge (up to 14,000 rpm)
- Pipettes and pipette tips
- 15 mL conical tube
- 1.5 mL microcentrifuge tubes
- Ethanol (96-100%)

6. Reagent Preparation

a) Prepare 90% Ethanol and 70% Ethanol Solutions:
 Add distilled water to the concentrated Ethanol (96-100%) (not supplied with ab156899) to obtain a 90% Ethanol Solution and a 70% Ethanol Solution.

b) Prepare DNU2/DNU3 Solution:

Add 1 mL of DNU2 to DNU3. Vortex until the solution is clear. The DNU2/DNU3 Solution can be stored at 4°C for up to 6 months.

7. Sample Preparation

Collect 5 ml of fresh urine into a 15 ml conical tube.

Note: If the urine sample will not be processed within a day of sample collection, protease inhibitors must be added.

Note: (1) About 6000 white cells may be expected per mL of urine in a healthy sample. Thus, the DNA yielded from 5 mL of urine would generally be a total of 50 to 100 ng or 2.5 to 5 ng/uL of eluate. (2) Using 0.5 to 1 uL of eluate is recommended for PCR analysis of urine DNA in order to reduce PCR inhibition due to possible co-eluted PCR inhibitors.

8. Assay Procedure

Note: Always close spin columns before placing them in the microcentrifuge.

- a) Centrifuge urine sample at 2000 rpm for 10 minutes to pellet cells.
- b) Remove supernatant and add 200 µL of DNU1 (Suspending Buffer) to suspend the cell pellet.
- c) Add 4 μ L of the mixed DNU2/DNU3 solution (see Section 6) to 200 μ L of cell suspension. Vortex and incubate at 65°C for 15 minutes. Meanwhile, place a spin column into a 2 mL collection tube.
- d) Add 300 µL of DNU4 (DNA Capture Buffer) to the cell suspension, mix, and transfer to the column. Spin for 45 seconds at 12,000 rpm. Discard the flowthrough. Replace the column to the collection tube
 - **Note:** *Maximum volume of the column is 600 μL.*
- e) Add 300 μL of 70% ethanol to the column and centrifuge at 12,000 rpm for 30 seconds. Discard the flowthrough and replace the column to the collection tube. Add 200 μL of 90%

- ethanol to the column and centrifuge at 12,000 rpm for 30 seconds.
- f) Discard the flowthrough and replace the column to the collection tube. Add an additional 200 μL of 90% ethanol to the column and centrifuge at 12,000 rpm for 40 seconds.
- g) Place the column in a new 1.5 mL vial. Add 8-18 μL of DNU5 (DNA Elution Buffer) directly to the column filter, and centrifuge at 12,000 rpm for 20 seconds to elute DNA. DNA is now ready for use or storage at -20°C.

9. Data Analysis

Typical Results

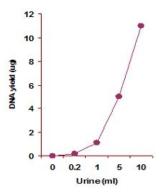


Fig. 1. Genomic DNA was isolated from Human urine using ab156899. The isolated DNA yield was quantified by real time PCR.



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