BioChain®
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User's Manual and Instructions

Urine DNA Isolation Kit

Catalog Number: K5011150

Shipping Condition: Room temperature

Introduction

The Urine DNA Isolation Kit is designed for the purification of genomic DNA and circulating DNA from urine samples. Purification is performed with the use of Spin columns which bind the DNA. No phenol-chloroform, protease, or precipitation steps are involved in this procedure. Isolation of the DNA is performed by lysing the cells in a solution containing chaotropic salts and detergent. Ethanol is added to the samples, which are then applied to the spin columns. Under these conditions the DNA binds to the membrane while other contaminants are washed through. The columns are then washed to further remove protein, buffer components and other contaminants using two ethanol-containing wash buffers and the final genomic DNA product is eluted in TE. The final DNA product can be used directly for quantitative PCR and other downstream applications.

Feature

- No phenol-chloroform
- No protease
- No precipitation
- Total <25 min.
- Sample range: 50 µl to 500 µl

Kit Contents

Item	Part #	Amount	Storage
1. Lysis Binding Buffer PC	K5011150-1	25 ml	RT
2. Wash Buffer 1	K5011150-2	14 ml	RT
3. Wash Buffer 2	K5011150-3	7 ml	RT
4. TE Buffer	K5011150-4	10 ml	RT
5. Spin column set	K5011150-5	50 units	RT

Special Handling instructions:

- Lysis Binding Buffer C contains hexadecyltrimethylammonium bromide and guanidine hydrochloride. Care should be taken in handling and disposal of samples.
- Wash Buffer 1 contains guanidine hydrochloride as a component. Care should be taken in handling and disposal of samples.

Storage Conditions

All of contents of the Urine DNA Isolation Kit including the buffers should be stored at room temperature. The kit is stable for one year under these conditions.

Technical Assistance

Please refer any technical questions to TechSupport@biochain.com.

Sample Size and Type

Urine DNA Isolation Kit can be used to isolate genomic DNA from fresh or frozen urine samples. If using frozen urine samples, thaw the samples on ice before starting the procedure.

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Buffer Concentrates
Wash buffers 1 and 2 are provided as concentrates that require the addition of 100% ethanol to them before use.

Reagents and Equipment to be Supplied by the User
• Pipettes
• 1.5 ml tubes
• Disposable gloves
• 100% ethanol
• Distilled deionized water
• A table-top centrifuge capable of providing >13k rpm rotor.

Before starting:

- Add 14 ml of 100% ethanol to Wash Buffer 1 and mark the bottle to indicate the addition of ethanol.
- Add 28 ml of 100% ethanol to Wash Buffer 2 and mark the bottle to indicate the addition of ethanol.

Protocol:

1. Transfer 500 µl of urine sample to a 1.5 ml tube.
2. Add 500 µl of Lysis Binding Buffer PC to the sample and mix well by vortexing. Allow the mixture to incubate at room temperature for 10 minutes.
3. After the incubation period, add 500 µl of 100% ethanol and mix well.
4. Apply 750 µl of the sample to a Spin column and centrifuge at full speed for 1 min.
5. Discard the flowthrough from the collection tube and reapply the Spin column onto the collection tube. Apply the remaining 750 µl of sample and centrifuge at full speed for 1 min.
6. Discard the flowthrough from the collection tube and reapply the Spin column onto the collection tube.
7. Add 500 µl of Wash Buffer 1 to the Spin column and centrifuge at full speed for 1 min. Ensure that ethanol has been added to Wash Buffer 1.
8. Discard the flowthrough. Add 600 µl of Wash Buffer 2 to the Spin column and centrifuge at full speed for 1 min. Ensure that ethanol has been added to Wash Buffer 2.
9. Discard the flowthrough and reapply the Spin column onto the collection tube. Dry the Spin column by centrifuging at full speed for 1 min.
   Place the Spin column onto a new 1.5ml microcentrifuge tube for elution. Apply 30-50 µl of TE Buffer to the center of the Spin column membrane and incubate for 1-5 min. Using a longer incubation results in more DNA recovery from the column. A lower volume of TE elution buffer will lead to a higher concentration of DNA. Using a higher volume of TE will result in higher recovery of DNA from the column.