

EpiQuik™ One-Step DNA Hydrolysis Kit

Base Catalog # P-1023

PLEASE READ THIS ENTIRE USER GUIDE BEFORE USE

The EpiQuik™ One-Step DNA Hydrolysis Kit is very suitable for digesting DNA from various biological materials. Before digestion, DNA should be purified and eluted in water. The digested DNA is suitable for multiple downstream applications including mass spectrometry, HPLC, and HPCE analysis.

The EpiQuik™ One-Step DNA Hydrolysis Kit allows DNA to be mainly digested into single nucleosides with very little of fragments having only 2-5 nucleosides.

KIT CONTENTS

| Components | 96 samples P-1023-96 |
|--------------------------|-------------------------|
| DH1 (Enzyme Mix) | 100 μ l |
| DH2 (Digestion Enhancer) | 100 μ l |
| DH3 (Digestion Buffer) | 5 ml |
| 96-Well PCR Plate | 1 |
| Adhesive Covering Film | 2 |
| User Guide | 1 |

SHIPPING & STORAGE

Upon receipt, DH1 and DH2 should be stored immediately at -20°C . All other components can be stored at room temperature. The kit is stable for up to 6 months from the date of shipment, when stored and handled properly.

MATERIALS REQUIRED BUT NOT SUPPLIED

- Waterbath or incubator
- Pipettes and pipette tips
- 1.5 ml microcentrifuge tubes

GENERAL PRODUCT INFORMATION

Product Updates: Epigentek reserves the right to change or modify any product to enhance its performance and design.

Usage Limitation: The *EpiQuik*[™] kits are for research use only and are not intended for diagnostic or therapeutic application.

Intellectual Property: *EpiQuik*[™] is a trademark of Epigentek Group Inc.

A BRIEF OVERVIEW

Digesting DNA to deoxyribonucleosides is a common process and a requirement for mass spectrometry and HPLC-based DNA analyses. These analyses include examination of the effects of gene polymorphisms and nutritional status on DNA metabolism, determination of the base composition of DNA, investigation of epigenetic modification, such as deoxycytosine methylation and oxidative damage. The currently used method for digesting DNA with tri-enzymes has several drawbacks, including: (1) a complicated procedure that includes adjusting pH twice, boiling samples, and separately incubating with different enzymes. This complicated procedure is time-consuming and limits the number of samples that can be prepared; and (2) the enzyme solution

used in such a method has high buffer concentrations that can interfere with the enzyme/DNA reaction and the downstream DNA analysis.

To address this issue, Epigentek offers the *EpiQuik*[™] One-Step DNA Hydrolysis Kit. This kit is designed for rapidly hydrolyzing DNA to deoxynucleosides with a process that can be performed in a single incubation. The *EpiQuik*[™] One-Step DNA Hydrolysis Kit has the following features:

- A fast one-step procedure, which can be finished in as short as 1 hour.
- Performed in a single incubation, without the need for DNA denaturation.
- Highly efficient enzymatic hydrolysis: non-specifically digests DNA or oligonucleotides into single nucleosides.
- 96-well microplate format makes the sample preparation flexible for use with high throughput formats.

PRINCIPLE & PROCEDURE

The *EpiQuik*[™] One-Step DNA Hydrolysis Kit simply applies our proprietary enzymatic DNA digestion solution to DNA or oligonucleotides. After treatment with the DNA digestion buffer, the DNA is easily digested into single nucleosides without phosphate groups.



Schematic Procedure for Using the *EpiQuik*[™] One-Step DNA Hydrolysis Kit

PROTOCOL

1. Isolate and purify DNA from cells/tissue or other biological materials using your own successful method. For your convenience and the best results, Epigentek offers a DNA extraction kit series (Cat. No.'s P-1003, P-1004, P-1017, and P-1018) optimized for isolation and purification of small scale DNA from tissue section, plasma/serum, urine, and blood/culture cells, respectively.
2. Measure DNA concentration and dilute DNA to 100-200 ng/ μ l with DNase/RNase free water.
3. Prepare the reaction mix (for 500-1000 ng of DNA). In each well, add:

| | |
|--------------|------------|
| DNA solution | 5 μ l |
| DH1 | 1 μ l |
| DH2 | 1 μ l |
| DH3 | 43 μ l |

Mix well by pipetting 2-3 times.

If the original DNA concentration is less than 100 ng/ μ l, the reaction mix can be prepared as the following:

| | |
|--------------|-------------|
| DNA solution | 5 μ l |
| DH1 | 0.5 μ l |
| DH2 | 0.5 μ l |
| DH3 | 14 μ l |

- Cover the plate with optical adhesive cover and incubate at 37°C (in a waterbath, incubator, or thermal cycler) for 1-2 hours.
- Incubate the samples at 95°C for 10 minutes to stop enzymatic reaction.
- If possible, take 2-5 μ l (50-100 ng DNA) of the digested samples, add loading buffer and run on agarose gel to check completeness of hydrolysis reaction. Make sure to run the un-digested DNA and DNA maker along with the digested DNA.
- Store the digested DNA at -20°C (for up to 6 months) or use immediately.

TROUBLESHOOTING

Un-Hydrolyzed or Incompletely Hydrolyzed DNA

Improper reaction conditions.

Check if the reaction conditions (temperature, time, amount of each component, etc.) are correct.

Too much DNA added into the reaction mix.

DNA amount added into the reaction mix should not be over 1000 ng.

Loss of enzyme activity due to improper storage conditions.

Check if the storage conditions of **DH1** and **DH2** are correct.

RELATED PRODUCTS

| | |
|--------|---|
| P-1003 | <i>FitAmp</i> [™] Tissue Section DNA Isolation Kit |
| P-1004 | <i>FitAmp</i> [™] Plasma/Serum DNA Isolation Kit |
| P-1017 | <i>FitAmp</i> [™] Urine DNA Isolation Kit |
| P-1018 | <i>FitAmp</i> [™] Blood and Cultured Cell DNA Extraction Kit |