TIANGEN®

# Hi-Swab DNA Kit

For purification of genomic DNA from buccal swab, throat swab, mouth wash, etc

# Hi-Swab DNA Kit

**(Spin Column)**

**Cat. no. GDP362**

## Kit Contents

| Contents | GDP362-01<br>50 preps | GDP362-02<br>200 preps |
|---|---|---|
| Buffer GHA | 30 ml | 120 ml |
| Buffer GBS | 15 ml | 60 ml |
| Buffer RD | 24 ml | 90 ml |
| Buffer PWE | 25 ml | 50 ml |
| Buffer TB | 15 ml | 30 ml |
| Proteinase K | 500 µl | 2 × 1 ml |
| Spin Columns CB2 | 50 | 200 |
| Centrifuge Tubes 1.5 ml | 50 | 200 |
| Collection Tubes 2 ml | 50 | 200 |
| Handbook | 1 | 1 |

## Optional Reagents

Swab storage buffer (TIANGEN)

## Storage

Hi-Swab DNA Kit should be stored dry at room temperature (15-30°C). Kit can be stored for up to 15 months without any reduction in performance and quality. If any precipitate forms in the buffers, it should be dissolved by warming the buffers at 37°C before use.

## Introduction

Hi-Swab DNA Kit uses unique silica membrane technology and special buffer system for purification of gDNA effectively. The Spin Columns CB2 made of new type silica-gel membrane in this kit can be easily and specifically bounded by DNA of many kinds samples such as buccal swab, pharyngeal swab and saliva. PCR inhibitors such as divalent cations and proteins are completely removed in two efficient wash steps, leaving pure DNA to be eluted in either distilled water (pH 7.0-8.5) or a buffer provided with the kit.

The genomic DNA isolated with the product possesses high quality and can serve as an excellent template for agarose gel analysis, restriction enzyme digestion, PCR and Southern blot.

## Important Notes Before starting

1. The samples should avoid repeated freezing and thawing, otherwise the extracted nucleic acid fragments will be smaller, and the extraction yield will be reduced.
2. If precipitates have formed in Buffer GBS, dissolve them by incubating at 37°C.
3. If the sample cannot be extracted in time, it can be stored in the Sample Preservation Buffer. Long-term preservation will not affect the extraction effect.

## Protocol

**Ensure that isopropyl alcohol has been added to Buffer GBS; while Buffer PWE been added with ethanol (96-100%) as indicated on the tag before use.**

1. Sample treatment

   1) Buccal swab sample extraction

      Transfer the buccal swab sample into the 2 ml centrifuge tubes and add 500 µl Buffer GHA. Add 10 µl Proteinase K, mix by vortex for 10 sec, and incubate at 65°C for 15-30 min. Mix every 10 min by vortex. Pipet out 300-350 µl reaction solution for the follow-up experiment.

   2) Pharyngeal swab sample extraction

      Transfer the pharyngeal swab sample into the 5 ml centrifuge tubes and add 1-2 ml Buffer GHA, mix by inverting upside down. Before extraction, pipet out 300-350 µl reaction solution and add 10 µl Proteinase K, mix by vortex for 10 sec, incubate at 65°C for 15-30 min and mix every 10 min by vortex.

   3) Saliva sample extraction

      Take the saliva sample as required, add equal volume Buffer GHA and mix by inverting upside down. Before extraction, pipet out 300-350 µl reaction solution and add 10 µl Proteinase K, mix by vortex for 10 sec, incubate at 65°C for 15-30 min and mix every 10 min by vortex.

      **Note: if the sample has been stored in the Sample Preservation Buffer of other manufacturers, add 0.5× volume Buffer GHA and 10 µl Proteinase K, mix by vortex for 10 sec, and incubate at 65°C for 15-30 min, pipet out 300-350 µl reaction solution for the follow-up experiment. If the Buffer GHA is not sufficient, it can be purchased separately.**

2. Add 500 µl Buffer GBS **(ensure isopropanol has been added)** to samples, mix well and incubate at room temperature(15-30°C) for 5 min.
3. Transfer all the lysate into CB2 (Placed in a 2 ml Collection Tube), close the lid, and centrifuge at 12,000 rpm (~13,400 x g) for 30 sec, then discard the filtrate and set the Spin Column CB2 back into the 2 ml Collection Tube.
4. Carefully open the Spin Column CB2 and add 500 µl Buffer RD **(ensure that ethanol (96-100%) has been added to Buffer RD before use)**. Close the lid and centrifuge at 12,000 rpm (~13,400 x g) for 30 sec, then discard the filtrate and set the Spin Column CB2 back into the 2 ml

Collection Tube

5. Repeat step 4.
6. Carefully open the Spin Column CB2 and add 600 µl Buffer PWE **(ensure that ethanol (96-100%) has been added to Buffer PWE before use)**. Close the lid and centrifuge at 12,000 rpm (~13,400 x g) for 30 sec, then discard the filtrate and set the Spin Column CB2 back into the 2 ml Collection Tube.
7. Repeat step 6.
8. Set Spin Column CB2 back into the 2 ml Collection Tube and centrifuge at 12,000 rpm (~13,400 x g) for 2 min. Discard the filtrate and incubate the Spin Column CB2 at room temperature (15-30°C) for several minutes to dry the membrane completely.
9. Place the Spin Column CB2 in a clean 1.5 ml microcentrifuge tube and pipet 50 µl Buffer TB on the center of the membrane. Close the lid and incubate at room temperature (15-30°C) for 2-5 min. Centrifuge at 12,000 rpm (~13,400 x g) for 2 min.
10. Collect the DNA into the tube and store in suitable condition.

## Determination of DNA Concentration and Purity

The recovered DNA fragments can be detected by agarose gel electrophoresis and ultraviolet spectrophotometer. DNA should have a significant absorption peak at $OD_{260}$. $OD_{260}$ value of 1 is equivalent to about 50 µg/ml double stranded DNA and 40 µg/ml single stranded DNA. The $OD_{260}$/ $OD_{280}$ ratio should be 1.7-1.9. If $ddH_2O$ is used for the elution instead of the elution buffer, the ratio will be lower, because the pH value and the presence of ions will affect the light absorption value, but it does not mean the purity is low.