

BW-MGD2318 Saliva gDNA Isolation Kit (Beads)

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Kit Contents

Catalog#	BW-MGD2318-A 00-00	BW-MGD2318-A0 0-01	BW-MGD2318-A0 0-02
Preps	50	250	1000
Swab Beads	0.6 mL	3 mL	12 mL
Buffer SBL	12 mL	60 mL	240 mL
Buffer MKB	36 mL	180 mL	3 x 240 mL
DNA Wash Buffer*	15 mL	72 mL	3 x 96 mL
Buffer MEB	6 mL	30 mL	120 mL
Proteinase K	1.2 mL	6 mL	24 mL
User Manual	1	1	1

*Add 60 mL (BW-MGD2318-A00-00) or 288 mL (BW-MGD2318-A00-01) or 384 mL (BW-MGD2318-A00-02) 96-100% ethanol to each DNA Wash Buffer bottle before use.

The **magnetic stand** needs to be purchased separately.

Introduction

Biomiga Saliva gDNA Isolation Kit provides a fast and simple procedure for isolating genomic DNA from saliva samples collected. Human genomic DNA extracted from buccal epithelial cells and white blood cells found in saliva can be used in various applications in diagnostics. The isolated DNA can be used for the detection of biomarkers to diagnose a disease, follow the diseases progress or monitor the effects of a particular treatment. Saliva DNA can also be used to diagnose particular types of infections. Isolation of DNA from saliva has become an attractive alternative to isolation from blood or tissue due to the fact that sample collection is non-invasive, the samples can be collected by individuals with little training, and no special equipment is required. Saliva gDNA purified using Biomiga kit is of the highest quality, and is compatible with a number of downstream research applications including PCR, Southern Blot analysis, sequencing and microarray analysis.

This kit is compatible with manual and automated processing.

Storage and Stability

Store Proteinase K and Swab Beads at 4°C upon receiving. From the date of production all other materials can be stored at room temperature (15-25°C). The guaranteed shelf life is 12 months from the date of production

Before Starting

Prepare all components and get all necessary materials ready by examining this user manual and become familiar with each step and pay special attention to the followings.

Important Notes

☼ Add 60 mL (BW-MGD2318-A00-00) or 288 mL (BW-MGD2318-A00-01) or 384 mL (BW-MGD2318-A00-02) 96-100% ethanol to each DNA Wash Buffer bottle before use.

☼ Prior to collection of saliva samples, the donor should rinse their mouth with a few milliliters of water for 10 seconds in order to remove any food particles that may be present. If food particles are present they may cause clogging of the column. 10 minutes after rinsing, collect saliva by spitting into a sterile collection tube or vital (not provided). The amount of saliva collected should be at least 100 µL but not more than 2 mL.

Materials not Supplied

- ☼ Sterile collection tubes, sterile microcentrifuge tubes.
- ☼ Water bath (55°C/65°C).
- ☼ Isopropanol.
- ☼ magnetic stand.

Safety Information

When working with chemicals, always wear a suitable lab coat, disposable gloves, and protective goggles. For more information, please consult the appropriate material safety data sheets.

Protocol

1. Sample pretreatment:

a. Fresh saliva sample

Collect 250 μ L fresh saliva sample into a sterile 2 mL centrifuge tube and add 250 μ L of Saliva Preservative.

Note: The Saliva Preservative needs to be purchased separately.

b. Saliva sample with protective solution

Collect 400-500 μ L saliva sample containing the protective solution (Saliva Preservative : Saliva = 1:1) into a sterile 2 mL centrifuge tube.

c. Oral swab sample

Add Saliva Preservative and PBS to 2 mL centrifuge tube (Saliva Preservative : PBS = 1:1), mix well, and the swab sample was cut and completely immersed in the liquid.

Note: Because the cotton swab absorbs liquid, make sure that the mixture has at least 400 μ L.

d. Oral swab sample with protective solution

Proceed directly to step 2.

2. Add **20 μ L Proteinase K** (vortex before use). Mix by vortexing and incubate at 55°C for 10-15 minutes.

3. Add **200 μ L Buffer SBL** to the saliva sample. Mix by vortexing and incubate at 55°C for 5 minutes.

4. Add 720 μ L Isopropanol and mix by vortexing for a few seconds.

5. Add **10 μ L Swab Beads** (vortex before use). Mix by vortexing and incubate at room temperature for 10 minutes, and mix once every 2 minutes.

6. Place the centrifuge tube on the magnetic stand for at least 1 minute, or until all the beads have collected. Keeping the tube on the magnet, carefully discard the supernatant with a pipette and take care not to attract the beads.

7. Remove the tube from the magnetic stand, then add **600 μ L Buffer MKB**, vortex and mix for 1 minute. Place the tube on the magnetic stand for 1 minute until the solution becomes clear.

Carefully discard the supernatant with a pipette and take care not to attract the beads.

8. Remove the tube from the magnetic stand, then add **600 μ L DNA Wash Buffer**, vortex and mix for 1 minute. Place the tube on the magnetic stand for 1 minute until the solution becomes clear. Carefully discard the supernatant with a pipette and take care not to attract the beads.
9. Repeat step **8**.
10. Try to remove the residual liquid with a pipette. Place the tube at room temperature for 5-10 minutes to dry the magnetic beads.
11. Add **50-100 μ L Buffer MEB** or ddH₂O, mix by vortexing and incubate at 65°C for 10 minutes.
12. Place the tube on a magnetic stand and let stand until the liquid is completely clear. Transfer all the supernatant to a sterile 1.5 mL centrifuge tube and store the collected DNA at -20°C.

Note: For automated DNA purification protocol or technical support, please contact us at 400-115-2855.

Trouble Shooting Guide

Problems	Possible Reasons	Suggested Improvements
The yield of genomic DNA is low	Swab Beads over dry	If the Swab Beads is too dry, re-clean the beads with 600 μ L of 75% ethanol.
	The Swab Beads did not break up fully	The Swab Beads must be thoroughly dispersed before use and during the extraction process.
	Incomplete lysis of cells	Increased Proteinase K incubation time at 55°C may result in increased yields
	The DNA elution is incomplete	Perform an additional centrifugation of 2 minutes at 14,000 rpm to ensure that all the DNA is eluted.
	DNA concentration in the saliva sample being used is low	Some saliva samples contain very little DNA. This varies from individual to individual based on numerous variables. Increased proteinase K incubation time at 55°C may result in increased yields.
DNA does not perform well in downstream applications	DNA was not washed with DNA Wash Buffer	Traces of salt from the binding step may remain in the sample if the column is not wash with DNA Wash Buffer. Salt may interfere with downstream applications, and thus must be washed from column.
	Ethanol carryover	Ensure that the dry spin after the column wash steps is performed, in order to remove traces of ethanol prior to elution. Ethanol is known to interfere with many downstream applications.

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RNA is present in eluted DNA	RNA is coeluted with the DNA	Carry out a digestion with RNase A on the elution if the RNase present will interfere with downstream applications. Refer to manufacturer's instructions regarding amount of enzyme to use, optimal incubation time and temperature.
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Limited Use and Warranty

This product is warranted to perform as described in its labeling and in BIOMIGA's literature when used in accordance with instructions. No other warranties of any kind, express or implied, including, without limitation, implied warranties of merchantability or fitness for a particular purpose, are provided by BIOMIGA. BIOMIGA's sole obligation and purchaser's exclusive remedy for breach of this warranty shall be, at the option of BIOMIGA, to replace the products. BIOMIGA shall have no liability for any direct, indirect, consequential, or incidental damage arising out of the use, the results of use, or the inability to use it product.

For technical support or learn more product information, please contact us at 400-115-2855 or visit our website at www.biomiga.com.cn