

Instructions for Isohelix Buccal Swab-Mag DNA Kit: BMG-12, BMG-50

Product Details

Isohelix buccal swab magnetic bead isolation kits have been specifically formulated to produce high DNA yield and purity from Isohelix buccal swabs, stabilised either with BuccalFix, Dri-Capsules, or RapiDri pouches. The kits have been fully optimised at Isohelix for use on buccal cell samples and offer reduced handling times, increased DNA yields and many other important technical benefits for their use in manual, or other high throughput formats.

Key Benefits

- ✓ Optimised for buccal cells
- ✓ Fast handling times
 - High purity and yield
 - ✓ Non-toxic reagents

- ✓ Protocol integrated to swabs
- Manual or high throughput formats
- ✓ No columns or filtration
- Less consumables wastage

Kit Contents

	Isohelix Buccal Swab-Mag DNA Kit for 12 or 50 buccal swabs		
Catalogue No.	BMG-12	BMG-50	Storage Temperature
Number of Buccal Swabs processed	12	50	
Contents:			
BLS Buffer (Lysis and Stabilisation buffer)	6ml	25ml	Room temperature
Proteinase K	2 x 2.2mg*1	1 x 22mg* <mark>2</mark>	4°C after reconstitution
Binding Buffer (BB)	6ml	25ml	Room temperature
Magnetic Beads (MB)	60µl	250µl	Store at 4 ^o C
Wash Buffer (MWB)	30ml* <mark>3</mark>	90ml*4	Room temperature
Elution Buffer (EB)	1.2ml	5ml	Room temperature
Protocol			

*1 Reconstitute each vial with 220µl sterile ddH₂O before first use, store at 4°C after reconstitution.

*2 Reconstitute vial with **1.1ml** sterile ddH₂O before first use, store at 4^oC after reconstitution.

*3 Add 21ml 98-100% Ethanol to the bottle before use.

*4 Add 63ml 98-100% Ethanol to the bottle before use.

Storage

Isohelix Buccal Swab-Mag DNA Kits are shipped at ambient temperature. <u>Please note</u> that on arrival the kit components should be stored according to the table above.

The kits are stable up to the expiry date if stored as instructed. See box label for expiry date.

Equipment and reagents to be supplied by user

- Waterbath or heating block at 60°C
- Pipettes with disposable tips
- Microcentrifuge (with rotor for 1.5 ml and 2.0 ml tubes)
- 2.0ml V bottom microcentrifuge tubes
- Vortexer
- 98-100% Ethanol, Molecular Biology Grade
- Magnetic Separation Rack compatible with 1.5ml/2.0ml microcentrifuge tubes, such as the Isohelix Mag-Rack (Cat No: MR-24).

Before Starting

- 1. Prepare waterbath or heating block at 60°C.
- 2. Reconstitute the Proteinase K by adding the appropriate amount of sterile ddH₂O as shown above.
- 3. Add the appropriate volume 98-100% ethanol to the Wash Buffer bottle as indicated above, invert to mix.
- 4. Vortex the tube of Magnetic Beads immediately prior to use, ensure the beads are all in suspension before adding to the sample.

Technical Assistance

If you have any questions regarding the use of this kit or other Isohelix products please contact us by email at <u>info@isohelix.com</u> or for further information visit the website at <u>www.isohelix.com</u>

Safety and Use of the Isohelix Buccal Swab-Mag DNA kits

Buffers in the Isohelix DNA kits contain irritants so appropriate safety equipment such as gloves, laboratory coats and eye protection should be worn. The kits are intended for use by qualified professionals trained in potential laboratory hazards and good laboratory practice. If direct information is not available on any of our compounds this should not be interpreted as an indication of product safety. This kit has been designed for Research Use Only.



Isolation Protocol:

Part A – DNA Stabilisation

Add 500µl BLS stabilisation buffer to the tube containing the swab, seal the tube then vortex briefly or invert to mix.
At this point the DNA is stabilised. You may continue with the DNA isolation or store the stabilised swab in a sealed tube at room temperature for at least 2 years. If swabs are stabilised in BuccalFix then this step can be skipped.

Part B - DNA Isolation

- 2. Add 20µl of reconstituted Proteinase K to the tube containing the buccal swab and BLS buffer. Replace the cap, vortex briefly and incubate @ 60°C for 30-60 minutes.
- 3. Transfer the liquid from the tube (approx. 400µl) into a 2ml V bottom microcentrifuge tube using a sterile pipette tip. *Optional step to increase yield:* Tip the swab head into a sterile 2ml centrifuge tube so that the swab head is uppermost. Spin the tube briefly and using a sterile pipette tip add the recovered supernatant to the 400µl collected previously.
- 4. Add 400μl Binding Buffer (500μl if using optional step 3), and 5μl fully resuspended magnetic beads to each tube. Vortex to mix.
- 5. Stand for 5 minutes at room temperature to allow the DNA to bind to the beads.
- 6. Place the uncapped tube in the magnetic rack using the six places sized for 2ml tubes, stand for 3 minutes to allow the beads to separate.
- 7. With the tube still in the rack, carefully remove all of the liquid using a 1ml pipette tip, and discard.
- 8. Transfer the tube to a separate rack. Add 1ml wash buffer and resuspend the magnetic beads either using a 1ml pipette tip or replacing the lid and vortexing the tube until all of the beads have been resuspended.
- 9. Replace the uncapped tube into the magnetic separator rack and leave for 2 minutes.
- 10. With the tube still in the rack, carefully remove all of the liquid using a 1ml pipette tip, and discard.
- 11. Repeat the wash steps 8-10 twice more, using 0.5ml Wash Buffer then 0.25ml Wash Buffer.
- 12. After the last wash has been removed, leave the uncapped tube in the magnetic rack for 5 minutes then use a pipette tip to remove any remaining liquid.
- 13. Add 100 μ l Elution Buffer, vortex to resuspend the beads. Incubate at 60°C for 2 to 5 minutes.
- 14. Replace the uncapped tube into the magnetic rack, leave for 3 minutes.
- 15. Using a pipette tip, carefully remove the eluate into a clean 2ml or 1.5ml microcentrifuge tube, taking care not to disturb the beads.
- 16. Check yield and purity by nanodrop. Generally, A260/280 >1.7, A260/230 >1.5. If levels are significantly lower than these, continue with step 18.
- 17. Centrifuge the sample for 5 minutes at 13,400 rpm/12,000 x g then carefully remove the supernatant to a clean 1.5ml or 2ml tube without disturbing any pellet.
- 18. Store the isolated DNA short term at 4° C or long term at -20° C or -80° C