Saliva Prep Xtra Kit for additional purification of GeneFix™ Saliva-Prep isolated samples: SPRE-50

Product Details

This kit is designed to further purify saliva DNA samples already isolated through GSP or GSPN Saliva-Prep kits, where the sample purity is below specified purity levels.

Kit Contents

Contents for processing 50 x 100µl DNA samples:		
Catalogue No.	SPRE-50	**Storage temperature**
TE buffer	15ml	Room temperature
Lysis buffer	10ml	Room temperature
Proteinase K	3 x 2.2mg*1	4^0C after reconstitution
SPN buffer	20ml	Room temperature

*1 Reconstitute each vial with **110µl** sterile ddH_2O before first use, store at 4^0C after reconstitution.

Storage

Isohelix GeneFix™ Saliva-Prep DNA Kits are shipped at ambient temperature.
Please note 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 60^0C
- Pipettes with disposable tips
- Microcentrifuge (with rotor for 1.5 ml and 2 ml tubes)
- 1.5ml or 2ml microcentrifuge tubes
- Vortexer

Before Starting

1. Prepare waterbath or heating block at 60^0C.
2. Reconstitute the Proteinase K by adding the appropriate amount of sterile ddH_2O as shown above.

Technical Assistance

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

Safety and Use of the Isohelix GeneFix™ Saliva DNA kits

Buffers in the GeneFix™ 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 practise. 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

Protocol for 100µl DNA sample previously isolated with GSP or GSPN kits

1. Add 100µl TE buffer to the 100µl DNA sample, vortex briefly. If the starting volume is less than 100µl, see **Note** below.
2. Add 200µl Lysis buffer, vortex briefly then add 5µl Proteinase K. Vortex briefly to mix.
3. Incubate at 60^0C for 15 minutes. Incubation can be extended to 1 hour or more.
4. Add 400µl SPN buffer, vortex well to mix thoroughly.
5. Centrifuge at maximum speed, 13.4K rpm/12,000 x g for 10 minutes.
6. Pour off the supernatant then re-spin briefly.
7. Remove all remaining liquid with a pipette tip taking care not to disturb the DNA pellet. It is important to remove all the liquid.
8. Add 100µl TE buffer to the pellet. Vortex well and leave at room temperature for at least 5 minutes for the DNA to re-hydrate.

Note: In step 1 if the starting volume is less than 100µl, add enough TE buffer to bring the total volume up to 200µl.
In step 8 if the starting volume was less than 100µl, resuspend the pellet in a volume of TE buffer equivalent to the starting volume.

Protocol for Isolation + Re-Extraction of GFX-02 4ml GeneFix™ saliva sample (2ml saliva collected into 2ml lysis buffer)

Part 1: GSPN isolation

1. Vortex the GeneFix™ saliva collection tube to mix well.
2. Add 40µl Proteinase K solution, vortex to mix then incubate at 60^{0}C for 30 minutes. Incubation can be extended to 1 hour or more.
3. Transfer the solution to a 15ml conical centrifuge tube. Add 4ml SPN buffer, vortex well to mix thoroughly.
4. Centrifuge at 4.4K rpm/3,000 x g for 30 minutes.
5. Pour off the supernatant then either stand upside down on absorbent paper to drain or re-spin briefly and remove all remaining liquid with a pipette tip taking care not to disturb the DNA pellet. **Note** it is important to remove all the liquid.
6. Add 500µl TE buffer to the tube, resuspend the pellet by pipetting or vortexing and leave at room temperature for at least 5 minutes for the DNA to re-hydrate.
7. Transfer the sample to a 2ml V-bottom microcentrifuge tube and centrifuge at maximum speed, 13.4K rpm/12,000 x g for 15 minutes to remove any undissolved particulates, remove supernatant to a clean 2ml microcentrifuge tube.

Part 2: SPRE re-extraction

8. Add 500µl lysis buffer to the 500µl DNA sample, then add 10ul Proteinase K and vortex briefly to mix.
9. Incubate at 60^{0}C for 15 minutes. Incubation can be extended to 1 hour or more.
10. Add 1ml SPN buffer, vortex well to mix thoroughly.
11. Centrifuge at maximum speed, 13.4K rpm/12,000 x g for 10 minutes.
12. Pour off the supernatant then re-spin briefly.
13. Remove all remaining liquid with a pipette tip taking care not to disturb the DNA pellet. It is important to remove all the liquid.
14. Add 400µl TE buffer to the pellet (or your preferred volume). Vortex well and leave at room temperature for at least 5 minutes for the DNA to re-hydrate.
15. If the sample still appears cloudy, centrifuge at maximum speed, 13.4K rpm/12,000 x g for 15 minutes to remove any undissolved particulates, remove supernatant to a clean 2ml microcentrifuge tube.