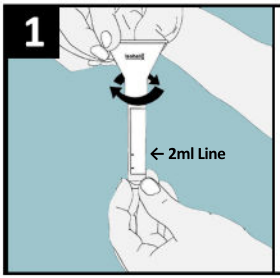
GeneFix

Patient Instructions for Saliva Collection

REF RFX-01

Do not eat, drink, smoke, brush your teeth or chew gum for 30 minutes before use.

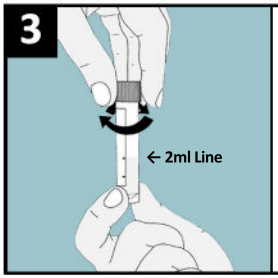
Check fluid level is at 1ml line before opening. If solution is cloudy, warm tube using hands for a few minutes.



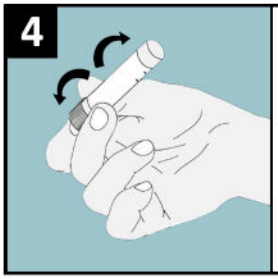
1. Remove the cap from the tube.
Keep cap for later use.
Screw collection funnel on to the tube.

2. Spit into the funnel until the level of the liquid in the tube, not including bubbles, **reaches the 2ml line.**

Delivery of saliva will take between 2 and 5 minutes.



3. Unscrew the collection funnel and replace the tube cap tightly.
Discard the funnel.



4. Shake the collection tube several times to mix the saliva with the solution.

Precautions: Do not swallow. If stabilising solution comes into contact with skin or eyes, wash with plenty of water. Tube cap may be a choking hazard to small children. MSDS available at www.isohelix.com.

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Version May 2020

CE 15°C 30°C

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Processing Instructions of RFX-01 saliva samples prior to RNA Isolation

Equipment & Reagents:

Supplied with kits:

- RFX Precipitation Reagent (store at room temperature).
- Proteinase K, 20 mg/ml (reconstitute lyophilised powder supplied with an appropriate volume of RNAse-free water prior to use, using the table below). Store reconstituted liquid at 4°C.

Proteinase K	Volume of H_2O to add
2.2 mg	110 µL
11 mg	550 µL
22 mg	1100 µL

To be supplied by user:

- RNAse-Free Water.
- Microcentrifuge tubes (1.5ml/2.0ml).
- Water/Dry Baths, preheated to 60°C and 90°C.
- Vortexer.
- Microcentrifuge (RCF ≥ 12,000 x g).

Processing Steps:

1. Gently vortex the saliva collection tube to mix. Remove a 300-500µl aliquot of sample into a clean, RNase-free 1.5ml or 2ml tube. The remainder of the RFX sample can be stored for later use at room temperature, or frozen for longer term storage at -20°C/-80°C.

2. Add 1/50 volume (6-10µl) of Proteinase K solution and vortex gently to mix.

3. Incubate aliquot at 60°C for 60 minutes, then heat @ 90°C for 15 minutes and cool to room temperature. Incubation time at 60°C can be reduced to 30 mins if required.

4. Add 1/25 volume (12-20µl) of RFX Precipitation Reagent. Vortex gently and incubate on ice for 10 minutes.

5. Spin at 12,000 x g for 3 minutes to pellet precipitate.

6. Carefully remove the supernatant into a clean, RNase-free 1.5ml or 2ml tube, taking care not to disturb the pellet. Discard the pellet.

7. Immediately add one volume of RNA lysis buffer from your RNA isolation kit of choice to the supernatant. Mix gently by inversion, then proceed with RNA isolation as per your isolation method protocol.