

## Supplementary Protocol for the Isolation of DNA from Norgen's Swab Collection and DNA Preservation System using Norgen's Saliva DNA Isolation Kit Product #: 45690

## Customer-Supplied Reagents

- Isopropanol
- 96-100% Ethanol
- Norgen's Swab Collection and DNA Preservation System (Cat# 45690)
- Norgen's Saliva DNA Isolation Kit (Cat# RU45400)

## Isolation of DNA from Swabs

- 1. Add 20 μL of **Proteinase K** (vortex before use) to the swab collection tube (blue cap). Mix by vortexing and incubate at 55°C for 1 hour.
- 2. Aliquot 500 µL of preserved sample to a microcentrifuge tube (user provided).
- 3. Add 200 µL of **Binding Buffer B** to the saliva sample. Mix by vortexing and incubate at 55°C for 5 minutes.
- 4. Add 720 µL of **Isopropanol** and mix by vortexing for a few seconds.
- 5. Assemble a column with one of the provided collection tubes.
- Apply up to 650 μL of the lysate to the Spin column and centrifuge for 1 minute at 6,000 RPM (~3,800 x g).

**Note:** Ensure the entire sample has passed through into the collection tube by inspecting the column. If the entire sample volume has not passed, spin for additional 1 minute at 10,000 RPM ( $\sim$ 10,000 x g).

- 7. Discard the flowthrough and reassemble the spin column with its collection tube.
- 8. Repeat with remaining lysate (you may need to repeat a total of three times to ensure all lysate passes through the column).
- Apply 500 μL of Wash Solution A to the column (ensure EtOH is added) and centrifuge for 1 minute at 8,000 RPM (~6,800 x g). Discard the flowthrough and reassemble the spin column with its collection tube.
- Apply 500 μL of Wash Solution A to the column and centrifuge for 1 minute at 14,000 RPM (~20,000 x g), a maximum speed. Discard the flowthrough and reassemble the spin column with its collection tube.
- 11. Spin the column for 2 minutes at 14,000 RPM (~20,000 x g) or at maximum speed in order to thoroughly dry the column. Discard the collection tube.
- 12. Place the column into a fresh 1.7 mL Elution tube provided with the kit.
- 13. Add 50 µL of Elution Buffer B to the column. Incubate for 5 minutes at 55°C.
- 14. Centrifuge for 2 minutes at 2,000 RPM (~ 425 x g), followed by a second spin for 1 minute at maximum speed.

## **Technical Support**

Contact our Technical Support Team between the hours of 8:30 and 5:30 (Eastern Standard Time) at (905) 227-8848 or Toll Free at 1-866-667-4362.

Technical support can also be obtained from our website (www.norgenbiotek.com) or through email at <u>techsupport@norgenbiotek.com</u>.

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