Saliva DNA Collection, Preservation and Isolation Kit – 50 Individual Devices

**Product Insert**

**Product #: RU35700**

Norgen’s Saliva DNA Collection, Preservation and Isolation Kit provides a safe and rapid all-in-one procedure for the collection, preservation and isolation of saliva DNA at ambient temperatures.

**INTENDED USE**

Norgen's Saliva DNA Collection, Preservation and Isolation Kit is an all-in-one solution designed for 1) simple and non-invasive saliva collection; 2) preservation of DNA in saliva samples at ambient temperature; and 3) isolation of high quality DNA within a laboratory setting. The Saliva DNA Collection, Preservation and Isolation Kit contains 50 Individual Saliva DNA Collection and Preservation Devices, as well as the required reagents for the subsequent laboratory isolation of the saliva DNA from the preserved samples. Each of the 50 individual Saliva DNA Collection and Preservation Devices consists of 3 components: (1) Saliva Collection Funnel and Collection Tube, (2) Collection Tube Cap, and (3) Norgen’s Saliva DNA Preservative contained within a sealed squeezable ampoule. Saliva samples are collected by spitting inside the Collection Funnel which has been assembled with the Collection Tube. After collecting the required volume of saliva the Collection Funnel is removed and the contents of the Preservative Ampoule are then added and mixed with the collected saliva. The Saliva Collection Tube is subsequently sent to the laboratory for DNA isolation and analysis. Each of Norgen’s Collection Tubes is labeled with a unique serial number that can be used for secure and anonymous tracking of the sample. The saliva DNA in preserved samples is stable for more than 5 years at room temperature. This kit is ideal for collecting, preserving and isolating DNA samples for epidemiological and population studies.

**SALIVA DNA PRESERVATIVE**

Norgen’s Saliva DNA Preservative is an aqueous storage buffer designed for rapid cellular lysis and subsequent preservation of DNA from fresh specimens. The buffer prevents the growth of Gram-negative and Gram-positive bacteria and fungi, and also inactivates viruses allowing the resulting non-infectious samples to be handled and shipped safely. In addition, the buffer eliminates the need to immediately process or freeze samples and allows the samples to be shipped to centralized testing facilities at ambient temperature. The components of the buffer allow samples to be stored for more than 5 years without any detectable DNA degradation.

**NUCLEIC ACID ISOLATION FROM PRESERVATIVE**

Saliva DNA can be isolated from the preserved saliva samples using one of four methods:

1- Alcohol precipitation using the provided reagents
2- Norgen’s spin-column format: Saliva DNA Isolation Kit (Cat# RU45400)
3- Norgen’s 96-well format: Saliva DNA Isolation 96-Well Kit (Cat # RU35200)
4- Norgen’s magnetic bead format: Saliva DNA Isolation Kit – Magnetic Bead System (Cat #RU55500)

**Saliva DNA Collection, Preservation and Isolation Kit Contents:**

<table>
<thead>
<tr>
<th>Component</th>
<th>Contents</th>
</tr>
</thead>
<tbody>
<tr>
<td>Individual Saliva DNA Collection and Preservation Device</td>
<td>50</td>
</tr>
<tr>
<td>Proteinase K</td>
<td>1.2 mL</td>
</tr>
<tr>
<td>Binding Buffer B</td>
<td>12 mL</td>
</tr>
<tr>
<td>Product Insert</td>
<td>1</td>
</tr>
</tbody>
</table>

**Individual Saliva DNA Collection and Preservation Device Contents:**

<table>
<thead>
<tr>
<th>Component</th>
<th>Contents</th>
</tr>
</thead>
<tbody>
<tr>
<td>Saliva Collection Funnel and Collection Tube</td>
<td>1</td>
</tr>
<tr>
<td>Collection Tube Cap</td>
<td>1</td>
</tr>
<tr>
<td>Preservative Ampoule</td>
<td>1</td>
</tr>
<tr>
<td>Donor Instructions</td>
<td>1</td>
</tr>
</tbody>
</table>
Kit Specifications

<table>
<thead>
<tr>
<th>Specification</th>
<th>Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Volume of Saliva Collected</td>
<td>2 mL</td>
</tr>
<tr>
<td>Volume of Saliva-Preservative Mix</td>
<td>4 mL</td>
</tr>
<tr>
<td>Preservation Temperature</td>
<td>Room temperature</td>
</tr>
<tr>
<td>Preservation Time</td>
<td>Over 5 years at room temperature</td>
</tr>
<tr>
<td>Time to Complete DNA Isolation</td>
<td>35 minutes</td>
</tr>
<tr>
<td>Average Yield from 0.5 mL*</td>
<td>5 μg - 20 μg</td>
</tr>
<tr>
<td>Average OD 260/280</td>
<td>1.7</td>
</tr>
</tbody>
</table>

* Average DNA yield will vary depending on the health status of the donor

MATERIALS REQUIRED BUT NOT SUPPLIED

For DNA Isolation
- Benchtop microcentrifuge (capable 20,000 x g)
- 55°C incubator
- Micropipettors
- 2 mL microcentrifuge tubes
- 15 mL centrifuge tube
- 70% ethanol
- Isopropanol
- TE Buffer (10 mM Tris pH 8.0, 1 mM EDTA)

SHELF LIFE AND HANDLING
- The Collection Device should be kept tightly sealed and stored at room temperature for up to 3 years without any reduction in kit performance (The collection due date is written on the device label).
- The kit contains ready-to-use Proteinase K which is dissolved in a specially prepared storage buffer. The Proteinase K should be stored at -20°C for up to 2 years after the “Collect Before Date” (with minimum freeze and thaw). The Proteinase K is a white turbid solution. A white precipitate is formed upon storage. **Make sure to re-suspend the white precipitate by vortexing before use.**
- Binding Buffer B should be kept tightly sealed and stored at room temperature. It can be stored for up to 2 years after the “Collect Before Date” without any reduction in kit performance.
- The Collection Tube, the Collection Funnel and the Device Container of each Individual Saliva DNA Collection and Preservation Device are recyclable.

QUALITY CONTROL
In accordance with Norgen’s Quality Management System, each lot of Norgen’s Saliva DNA Collection, Preservation and Isolation Kit is tested against predetermined specifications to ensure consistent product quality.

Disclaimers and Satisfaction Guarantee
For research use only. Not for use in diagnostic procedures.

NORGEN BIOTEK CORPORATION guarantees the performance of all products in the manner described in our product manual. The customer must determine the suitability of the product for its particular use. We reserve the right to change, alter, or modify any product to enhance its performance and design.

WARNINGS AND PRECAUTIONS – Saliva Donor
1. If any of the Saliva DNA Preservative comes into contact with skin or eyes, wash thoroughly with water. Do not ingest the Saliva DNA Preservative.
2. The collection device contains a choking hazard (Collection Tube Cap). Keep away from children.
WARNINGS AND PRECAUTIONS – DNA Isolation from Preserved Saliva Samples

1. Follow universal precautions. All specimens should be considered as potentially infectious and handled accordingly.
2. Wear personal protective equipment, including gloves and lab coats when handling kit reagents. Wash hands thoroughly when finished performing the procedures.
3. Do not smoke, drink or eat in areas where kit reagents and/or human specimens are being used.
4. Dispose of unused kit reagents and human specimens according to local, provincial or federal regulations.
5. As contamination of specimens or reagents can produce erroneous results, it is essential to use aseptic techniques.
6. Only use the protocol provided in this insert. Alterations to the protocol and deviations from the times and temperatures specified may lead to erroneous results.
7. For more information, please consult the appropriate Material Safety Data Sheets (MSDSs). These are available as convenient PDF files online at www.norgenbiotek.com.

Procedures

A. SAMPLE COLLECTION AND PRESERVATION PROCEDURES

1. Do not eat, chew gum, drink or smoke for 30 minutes prior to saliva collection.
2. Rinse your mouth with water and wait for 10 minutes.
3. Spit into the Collection Funnel until the amount of liquid saliva (not including bubbles) in the Collection Tube reaches the 2 mL line. Discard / recycle the Collection Funnel.
4. Twist and remove the tip of the Preservative Ampoule.
5. Squeeze the Preservative Ampoule to empty the contents into the Collection Tube. Screw on the cap of the Collection Tube to close it tightly.
6. Shake the Collection Tube well for 10 seconds to mix saliva and preservative.
7. The saliva sample is now preserved and ready for storage, shipping or processing.
   **Note:** Each of Norgen’s Collection Tubes is labeled with a unique serial number that can be used for secure and anonymous tracking of the sample.
8. Dispose of the Preservation Ampoule and tip in the garbage. The plastic device container can be recycled.

B. INSTRUCTIONS FOR STORAGE OF PRESERVED SALIVA SAMPLES

Preserved saliva samples can be stored at room temperature for more than 5 years without significant loss of DNA quality.
Flowchart
Procedure for Isolating Saliva DNA from 0.5 mL of Preserved Saliva Samples

Mix preserved saliva sample and transfer 0.5 mL

Add Proteinase K, mix. Incubate at 55°C for 10 minutes
Add Binding Buffer B, mix. Incubate at 55°C for 5 minutes

Add isopropanol and mix

Centrifuge for 3 minutes.

Carefully remove and discard supernatant. Add 70% ethanol. Stand 1 minute.

Centrifuge for 1 minute.

Carefully remove and discard supernatant. Rehydrate in TE Buffer

Incubate at 55°C for 5 minutes.

Pure Saliva DNA
C. ISOLATION OF SALIVA DNA FROM 0.5 mL OF PRESERVED SALIVA SAMPLES

Note: The amount of Proteinase K and Binding Buffer B provided are sufficient for 50 isolations from 0.5 mL of preserved saliva. For DNA isolation from larger volumes of preserved saliva (up to 4 mL) you can purchase Norgen’s Saliva DNA Isolation Reagent Kit (up to 4 mL) (Cat# RU35720).

1. Mix the preserved saliva sample by inversion and gentle shaking for a few seconds.
2. Transfer 0.5 mL of preserved saliva sample to a 2 mL centrifuge tube (not provided).
3. Add 20 µL of Proteinase K (make sure to vortex proteinase K before use), mix by vortexing for 10 seconds and incubate the sample at 55°C for 10 minutes.
4. Add 200 µL of Binding Buffer B to the sample. Mix by vortexing for 10 seconds and incubate the sample at 55°C for 5 minutes.
5. Add an equal volume (total volume of preserved saliva, Proteinase K and Binding Buffer B) of room temperature isopropanol to the sample. Mix gently by inversion 10 times.
6. Centrifuge at room temperature for 3 minutes at maximum speed at 20,000 x g (~14,000 RPM).
7. Carefully remove and discard the supernatant, taking care not to disturb the DNA pellet. Gently place the tube briefly upside down on a paper towel to remove residual isopropanol.
8. Carefully add 500 µL of 70% ethanol. Gently swirl and let stand at room temperature for 1 minute.
9. Centrifuge at room temperature for 1 minute at maximum speed 20,000 x g (~14,000 RPM) then carefully remove and discard the ethanol without disturbing the pellet.
10. Gently place the open tube upside down on paper towel for 5 minutes to remove the excess amount of 70% EtOH and to air dry the DNA pellet.
11. Add 100 µL of TE Buffer. Vortex for 30 seconds and incubate at 55°C for 5 minutes to rehydrate the DNA pellet. Ensure complete rehydration of the DNA (pellet and smear on the side of the tube) prior to any subsequent steps.

Note:
   a. If a higher DNA concentration is required for downstream applications, the DNA pellet can be rehydrated in 50 µL of TE Buffer.
   b. Large amounts of high molecular weight DNA can be very slow to rehydrate (dissolve) completely. Incomplete rehydration of the DNA will affect estimating the DNA concentration and can result in failure of downstream applications such as SNP, digestion and PCR.
12. Centrifuge the tube at 20,000 x g (~14,000 RPM) for 1 minute to pellet any insoluble material.
13. Transfer the clear liquid into a clean tube taking care not to disturb the pellet.
14. The purified DNA sample may be stored at 4°C up to 2 months. It is recommended that samples be placed at ~20°C for long term storage.
**Related Products**

<table>
<thead>
<tr>
<th>Product Description</th>
<th>Product #</th>
</tr>
</thead>
<tbody>
<tr>
<td>Saliva DNA Isolation Reagent Kit (up to 4 mL)</td>
<td>RU35720</td>
</tr>
<tr>
<td>Shipping Accessories – 50 Mailers</td>
<td>38300</td>
</tr>
<tr>
<td>Saliva DNA Isolation Kit</td>
<td>RU45400</td>
</tr>
<tr>
<td>Saliva DNA Collection and Preservation Devices (50)</td>
<td>RU49000</td>
</tr>
<tr>
<td>Saliva DNA Isolation Kit (Magnetic Bead System) – 50 Preps</td>
<td>RU55500</td>
</tr>
</tbody>
</table>

**Technical Assistance**

NORGEN’s Technical Service Department is staffed by experienced scientists with extensive practical and theoretical expertise in sample and assay technologies and the use of NORGEN products. If you have any questions or experience any difficulties regarding Norgen’s Saliva DNA Collection, Preservation and Isolation Kits or NORGEN products in general, please do not hesitate to contact us.

NORGEN customers are a valuable source of information regarding advanced or specialized uses of our products. This information is helpful to other scientists as well as to the researchers at NORGEN. We therefore encourage you to contact us if you have any suggestions about product performance or new applications and techniques.

For technical assistance and more information, please 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 or call one of the NORGEN local distributors ([www.norgenbiotek.com](http://www.norgenbiotek.com)) or through email at techsupport@norgenbiotek.com.