Isolation of High Quality Saliva DNA from Variable Donor Saliva Samples

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INTRODUCTION

In recent years attention has been turning to the use of non-invasive samples for genetic and diagnostic analysis, including the use of saliva. In contrast to blood samples, saliva can be self-collected, is less costly to ship and easier to store and process. Human genomic DNA extracted from buccal epithelial cells and white blood cells found in saliva can be used in various applications including diagnostic assays, epidemiological studies and surveys.

The isolation of high quality DNA from saliva is not without its problems however. The number of DNA-containing cells found in saliva can vary significantly from individual to individual based on a number of different factors, including health status of the individual donor. Adequate amounts of saliva must therefore be collected to ensure that DNA can be extracted in an amount sufficient for testing. This application note reports on the integrity and size of saliva DNA isolated from 60 different donors using Norgen's Saliva DNA Collection, Preservation and Isolation Kit (Cat #35700).

METHODS AND MATERIALS

DNA Isolation

Sixty different saliva samples were collected using Norgen's Saliva DNA Collection, Preservation and Isolation Kit (Cat# 35700). DNA was purified according to the supplied protocol. An additional 55°C incubation step was carried out for one hour in order to fully rehydrate the isolated DNA.

Gel electrophoresis

For visual analysis of DNA size and integrity, 10 µL of DNA from the final DNA elution was loaded on a 1% agarose TAE gel and run for 25 minutes at 150 V. Gel photo was taken by AlphaImagerTM IS-2200 (Alpha Innotech).

DNA Sequencing and analysis

Three of the PCR products were first cleaned using Norgen's PCR Purification kit (Norgen Biotek, Cat# 14400) and sent to University of Guelph Laboratory Service for sequencing (Guelph. ON). Using forward and reverse primers, the PCR product was sequenced by ABI Prism 3730 & 3100 DNA sequencers. The sequencing data was analyzed by Sequence Scanner software v 1.0 (Applied Biosystems).

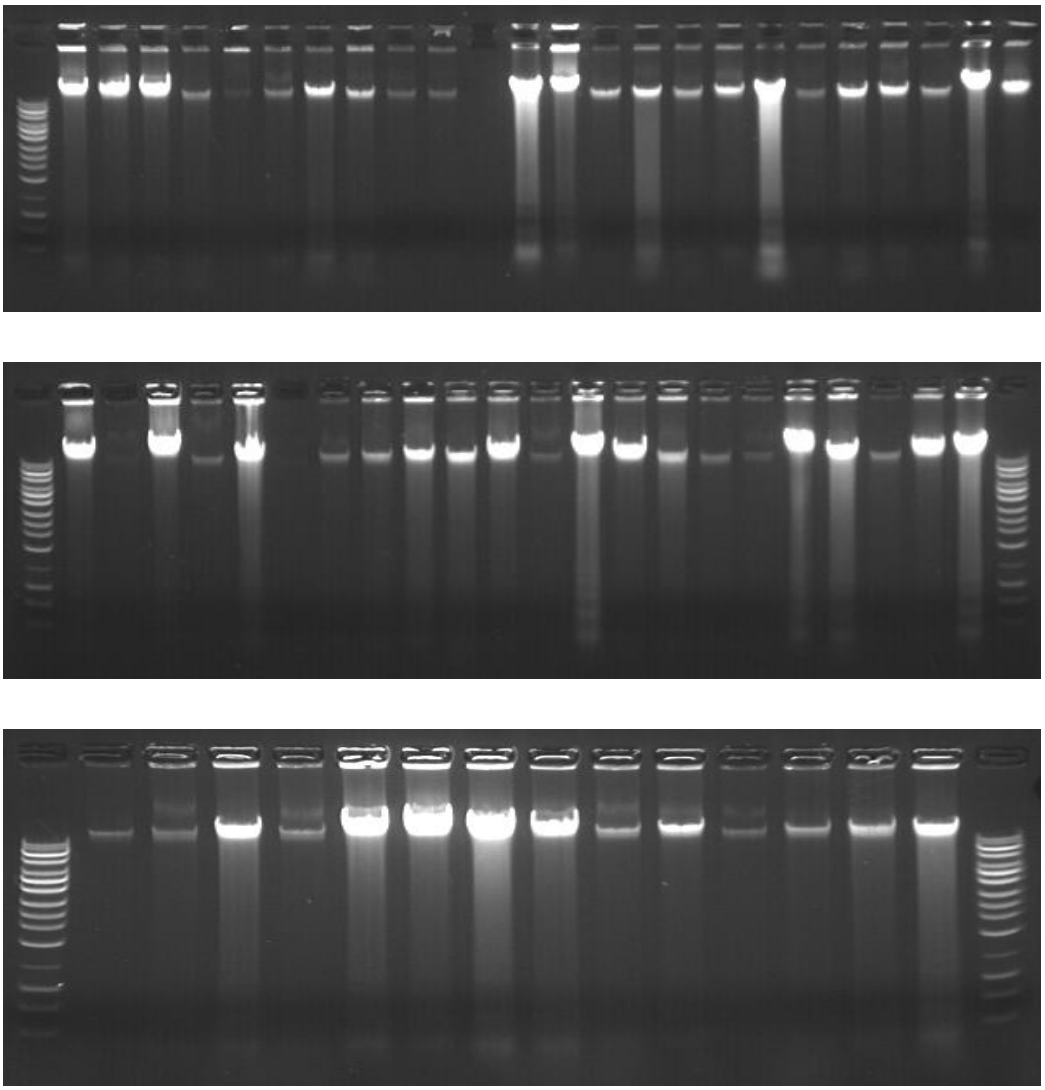

Figure 1. Total DNA isolated from 60 variable donors. The 1% agarose TAE gel shows the different ranges of DNA yield, as well as the high molecular weight of the isolated DNA. Norgen's UltraRanger 1 kb DNA Ladder (Cat# 12100) was used.

RESULTS AND DISCUSSION

In this application note, 60 saliva samples were collected from 60 different donors using Norgen's Saliva DNA Collection, Preservation and Isolation Kit. Total DNA wasthen isolated from 0.5 mL of saliva/preservation mix using the provided protocol. The purified DNA was loaded on a 1% agarose gel in order to visually analyze DNA integrity and size **(Figure 1)**.

The gel picture in **Figure 1** demonstrates the variation of saliva DNA yield isolated from different individual donors. Despite variability between samples from the 60 different donors, Norgen's Saliva DNA Collection, Preservation and Isolation Kit successfully isolated high quality DNA without any evidence of degradation. Furthermore, the size of the isolated DNA was over 24 kb in all cases, as evidenced by the fact that all samples are over the top band of the marker used, which is 24 kb (Norgen's UltraRanger 1 kb DNA Ladder, Cat# 12100).

CONCLUSIONS

Sixty saliva samples from individual donors were successfully isolated using Norgen's Saliva DNA Collection, Preservation and Isolation Kit. The gel electrophoresis results reveals that the saliva DNA isolated using Norgen's Saliva DNA Collection, Preservation and Isolation Kit is of a high quality regardless the personal variability.

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