Comparison of Norgen's Saliva DNA Collection, Preservation and Isolation Kit to a Leading Competitor's Kit

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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. Norgen Biotek Corp. has developed a simple method for the collection, preservation, storage and purification of DNA from saliva using Norgen's Saliva Collection, Preservation and Isolation Kit (Cat #35700). Saliva samples collected with this kit are stable at room temperature for over 2 years in the provided Saliva DNA Preservative. The resulting purified DNA is of the highest quality and can be used in a number of downstream applications. In this application note 10 different saliva samples were collected using Norgen's Saliva DNA Collection, Preservation and Isolation Kit. The DNA was subsequently isolated and then used successfully for sequencing.

METHODS AND MATERIALS

DNA Isolation

Ten 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.

Amplification of by PCR

One hundred nanograms of each of the isolated DNA samples were used as the template in an end-point PCR reaction using 2X PCR Master Mix (Norgen Biotek). Human Thymidylate Synthase specific primers (forward: 5' ATGCTTAGTAGGCAATTCTG3'; reverse: 5' TTTGGTTGTCA GCAGAGG 3', amplicon size of 560bp were used in the reaction amplified on the iCycler iQ real-time system (BioRad).

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).

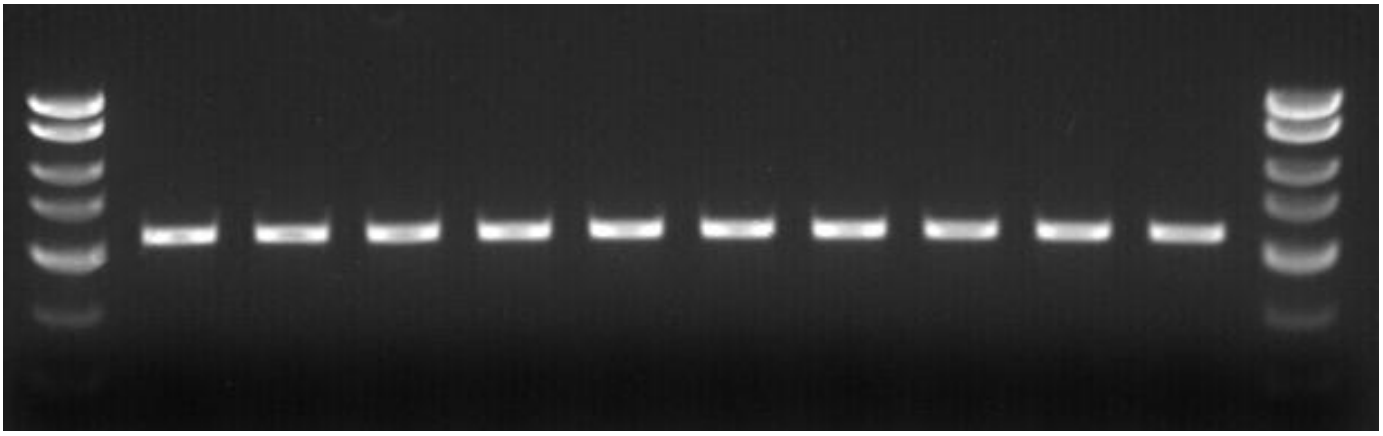

Figure 1. PCR amplification of 10 saliva DNA samples isolated using Norgen's Saliva DNA Collection, Preservation and Isolation Kit.

RESULTS AND DISCUSSION

PCR is a common downstream application that indicates DNA quality. For this application note, 10 different saliva samples were collected and processed using Norgen's Saliva DNA Collection, Preservation and Isolation Kit. All of the ten saliva DNA samples were successfully amplified in a PCR reaction using Human Thymidylate Synthase specific primers (Figure 1), indicating that no PCR inhibitors were co-purified with the DNA.

The PCR products were cleaned using Norgen's PCR Purification Kit (Cat# 14400) prior to sending for sequencing analysis. As can be seen in Figure 2 all bases were accurately read and generated clean sequencing reading profile without sequencing interruption.

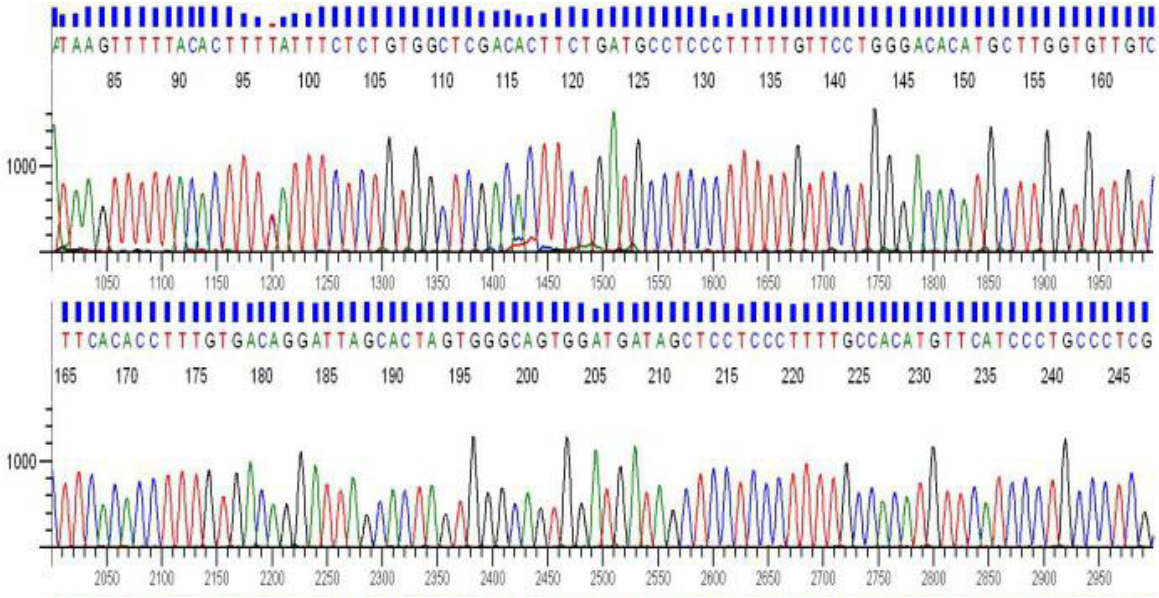

Figure 2. Sequencing reading from ABI Prism 3730 sequencer.

CONCLUSIONS

Norgen's Saliva DNA Collection, Preservation and Isolation Kit provides high quality saliva DNA that is compatible with PCR and sequencing analysis without showing PCR inhibition or interruption of sequencing reading.

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PCR Purification Kit	50 Preps	14400

For more information
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