Isolation of high purity, high quality DNA from buccal swabs using the Isohelix™ Buccal-Prep Plus Kit (BPP-50)

Introduction

The aim of this investigation is to demonstrate the potential of the new Isohelix™ Buccal-Prep Plus. This addition to the Isohelix™ family provides significant improvements to the purity of isolated buccal samples, precipitating out impurities while maximising recovery of DNA. This application note demonstrates the ability of this improved kit to isolate DNA of high quality and purity from Isohelix™ buccal swabs.

Methods & Materials

Ten buccal swab samples were taken by a number of adult volunteers using SK-1 buccal swabs. The swabs were used for one minute, following which the swab heads were snapped off into the 5ml tubes provided. The swabs were then stabilised, then isolated using the Isohelix™ Buccal-Prep Plus, following the kit's protocol.

The purified samples were tested for DNA concentration and yield using the Qubit BR dsDNA assay, whilst purity of samples was assessed through Nanodrop-1000 absorbance ratio analysis. Samples were also visually analysed via agarose gel electrophoresis.

Results

Table (1): Qubit dsDNA BR Assay & Nanodrop data of buccal swab samples isolated using the Buccal-Prep Plus isolation Kit, displaying sample yield and purity.

	Qubit dsDNA BR Assay Data		Nanodrop Data		
Sample	**Sample Concentration (µg/ml)**	**Total DNA Yield (µg)**	**A260/280**	**A260/230**	**Sample Concentration (µg/ml)**
1	23.40	2.34	1.80	1.62	27.78
2	29.00	2.90	1.77	1.29	36.85
3	24.90	2.49	1.88	1.31	30.89
4	43.00	4.30	1.77	1.50	52.73
5	74.40	7.44	1.78	1.73	93.39
6	45.30	4.53	1.79	1.67	55.67
7	57.30	5.73	1.76	1.69	57.61
8	125.00	12.50	1.76	1.49	164.89
9	31.00	3.10	1.84	1.30	40.96
10	36.00	3.60	1.77	1.27	40.24
Mean	**48.93**	**4.89**	**1.79**	**1.49**	**60.10**

In this investigation, the yields that were obtained from the buccal swabs ranged from 2.34-12.50 µg, with a mean yield of 4.89 µg, and a mean concentration of 48.93 µg when measured by Qubit. A260/280 ratios ranged from 1.76-1.88 with a mean value of 1.79. A260/230 ratios ranged from 1.27-1.73 with a mean value of 1.49. Concentrations recorded by Nanodrop ranged from 27.78-164.89 µg, with a mean concentration of 60.1 µg.

To further analyse the quality of DNA isolated, the samples were run on a 2.2% Agarose FlashGel™ to check the integrity of the whole DNA and for the presence of any RNA in the samples against a 100-4000bp ladder.

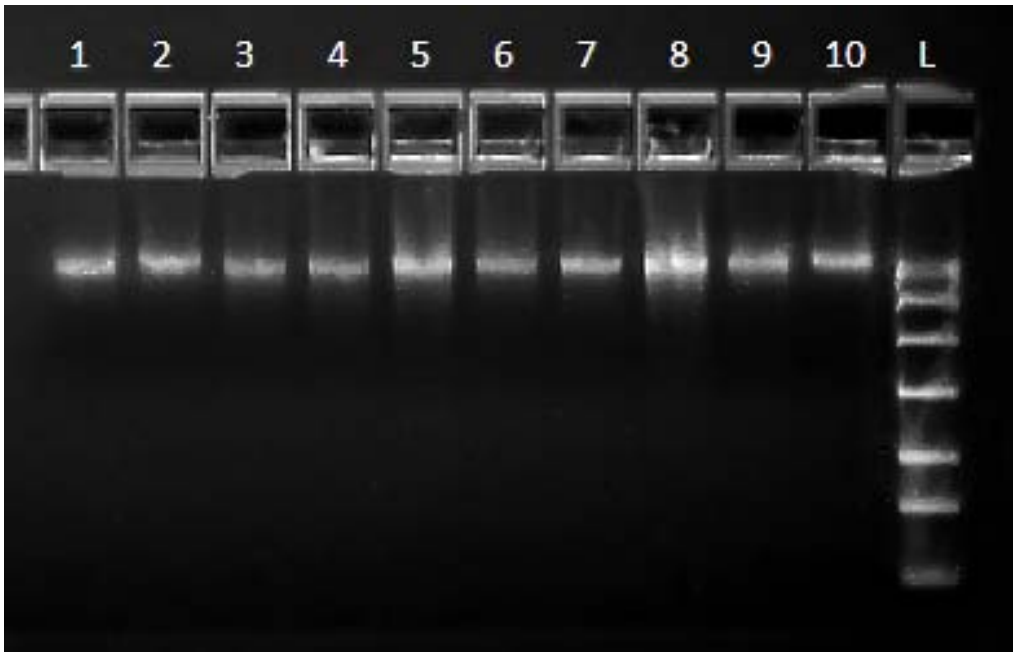

Figure (1): 2.2% Agarose Flash Gel of whole DNA samples (Diluted to 1/10) isolated using the Isohelix Buccal-Prep Plus kit from SK buccal swabs.

From the gel samples run (Figure 1) it can be seen that all swabs isolated show high molecular weight genomic DNA with no signs of fragmentation or shearing. In addition to this no RNA contamination is seen.

In conclusion, these results show us that the new Isohelix™ Buccal-Prep Plus DNA Isolation Kit can reliably purify high quality DNA from a range of buccal swab samples with no fragmentation and without co-elution of RNA.