Estimating Human and Bacterial DNA Content in GeneFix Saliva Samples

To estimate relative quantities of Human and microbial DNA found in GeneFix stabilised samples, saliva samples from ten donors (labelled A through J) were collected into GeneFix GFX-02 saliva collectors, which were then stored at room temperature prior to DNA extraction. The saliva samples were then purified using the GeneFix Saliva-Prep DNA Isolation Kit (GSPN), following which they were analysed for yield & purity by spectrophotometer and fluorometric assays.

Using the quantification data, DNA concentrations were normalised to approximately 10ng/µl, verified using the fluorometric assay. The normalised DNA samples were then analysed using qPCR to determine relative human and microbial content in each.

Two separate qPCR assays were performed using the same set of normalised samples. The first targeted the Human ACTB gene, while the second targeted the Bacterial 16S rRNA gene. Test samples were run alongside a standard curve generated using either pure Human or bacterial DNA for each respective assay, which was used to estimate relative human/microbial content of samples and to determine qPCR efficiency. Percentage DNA estimates were determined by dividing these values against the concentrations determined using the fluorometric assay previously.

Results & Conclusions:

	% Human DNA Estimate	% Bacterial DNA Estimate
A	67.7	14.8
B	79.8	4.4
C	85.5	2.1
D	82.5	6.5
E	76.3	27.2
F	89.3	2.8
G	72.4	17.2
H	77.7	8.4
I	71.1	11.5
J	101.1	0.9
Median	**78.76**	**7.43**

- **On average, the majority of DNA derived from saliva samples stabilised using GeneFix collectors was estimated to be of Human origin (median 78.76%) as determined by qPCR, with a smaller percentage of DNA estimated to be of microbial origin (median 7.43%).**

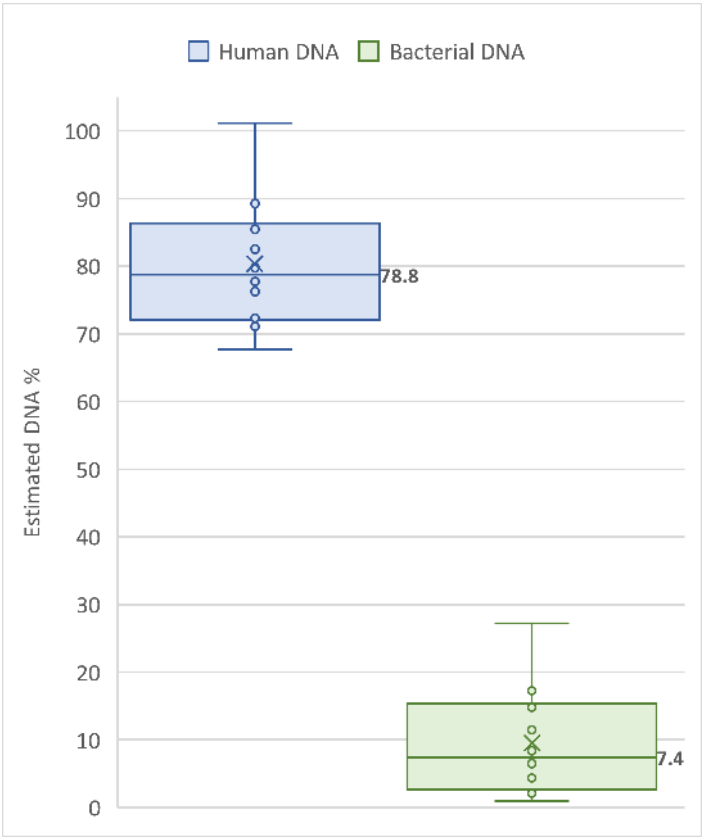

Figure 1: Box plot detailing relative percentages of human and microbial DNA content found in a set of GFX-stabilised saliva samples.