Smarter DNA Collection devices

An investigation into the yields of Isohelix Buccal swabs compared with other commercially available swabs.

Introduction

The aim of this paper is to demonstrate the suitability of Isohelix buccal swabs as an alternative to venopuncture for the collection of nucleic acids for use in diagnostic testing and their superior yield over other commercially available swabbing devices.

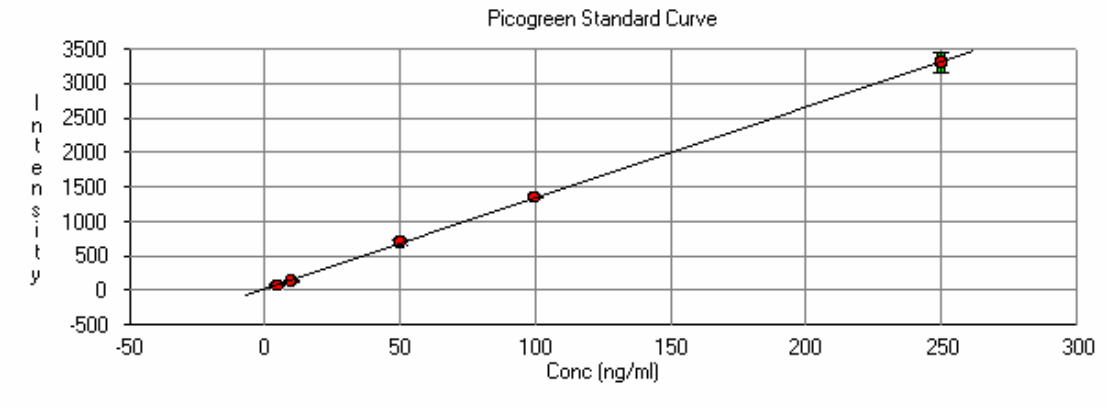
The most common source of nucleic acids for use in Diagnostic and Research laboratories is blood, which gives good yields of nucleic acids which is ideal for use in many modern tests where multiple assays are necessary (SNP analysis, Haplotyping etc). The use of blood for these assays is not without problems, collection requires the use of a healthcare professional qualified to carry out venopuncture that can be costly, there are issues over the transport of blood and blood based samples. Buccal cell swabs are often used as an alternative to blood collection, it is cheap, painless and can easily be done by the subject, however the problem with buccal swabs has been relatively low yields.

Experiment

The Isohelix swab was compared with two other commercially available swabs. The first (***swab A***) is a compressed paper product, the paper head is held on a tubular plastic handle, the second (***swab B***) had a foam head that is glued on to a solid plastic handle.

A total of nine samples were taken using each of the three swabs, from nine different individuals on three separate days (one swab type per day). One unused swab of each type was isolated as a control to check for contamination. Genomic DNA was isolated from all of the swabs using the Qiagen Qiamp (1) DNA mini kit following a standard protocol (2); the Genomic DNA was eluted in a volume of 200 ul. The DNA isolates were then quantified using the Molecular Probes Picogreen dsDNA quantification reagent (3), A standard curve was prepared using Lambda DNA with a final concentration range of 5, 10, 50, 100 and 250 ng/ul , in order for the results of this assay to be acceptable an R^2 of 0.995 or greater was necessary from the standard curve. The plate was prepared in accordance with a standard protocol ad the standards, no DNA controls and DNA isolates were pipetted into a plate containing the Picogreen reagent. The plate was then read using a Biotek FL606F fluorescence microplate reader (excitation 480 nm, emission 520 nm)(4)

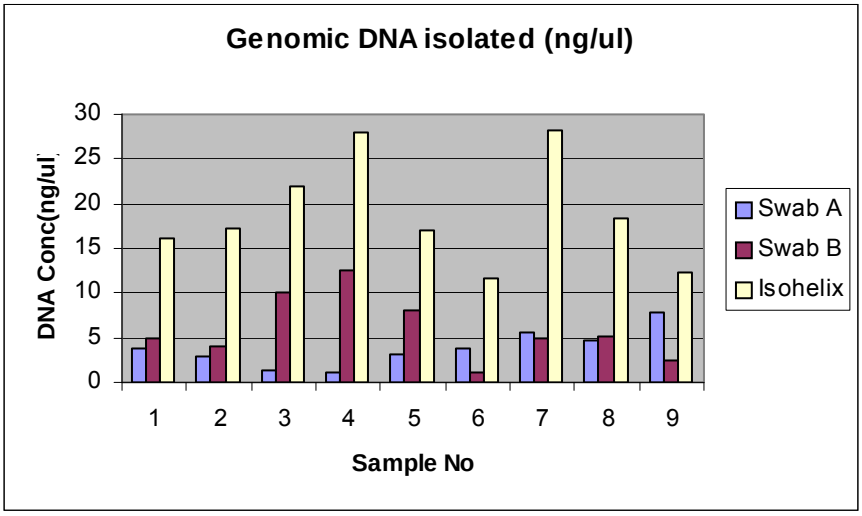
Results



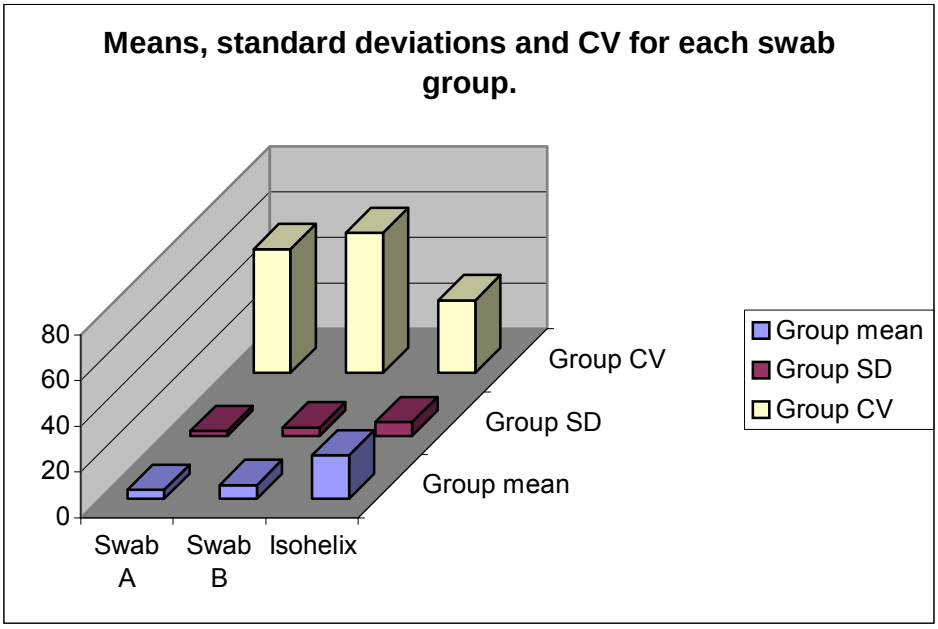
Standard curve, linear regression Y =a*x+b, a=13.20, b=18.10, R=0.9999, R^2=0.9998, err=17.34

Genomic DNA concentrations determined by the picogreen assay

Sample No	Swab A	Swab B	Isohelix
1	3.9162 ng/ul	5.0123 ng/ul	16.203 ng/ul
2	2.9917 ng/ul	4.0716 ng/ul	17.253 ng/ul
3	1.3246 ng/ul	10.133 ng/ul	21.872 ng/ul
4	1.1882 ng/ul	12.529 ng/ul	27.959 ng/ul
5	3.1736 ng/ul	7.9703 ng/ul	16.903 ng/ul
6	3.848 ng/ul	1.1819 ng/ul	11.607 ng/ul
7	5.5606 ng/ul	4.8377 ng/ul	28.233 ng/ul
8	4.7429 ng/ul	5.1287 ng/ul	18.25 ng/ul
9	7.9049 ng/ul	2.5586 ng/ul	12.231 ng/ul
10 (blank)	0 ng/ul	0 ng/ul	0 ng/ul



Swab type	Group mean (ng/ul)	Group Standard Deviation	Group CV
Swab A	3.85	2.087	54.195
Swab B	5.93582	3.63	61.214
Isohelix	18.9457	6.02	31.782



Conclusion.

As the experimental daa clearly demonstrates the Isohelix swab performs significantly better than other Buccal cell collection swabs available in the market, based upon the group mean of 18.947 ng/ul the total DNA recovered from the Isohelix swab is 3.79 µg of genomic DNA, compared to 770 ng for swab A and 1.18 µg for swab B, the yield of 3.79 µg gives sufficient Genomic DNA to carry out over 150 Taqman quantitative PCR reactions (5) with a 25 µl reaction volume. This makes the use of this swab for haplotyping and SNP genotyping tests and studies a viable, cost effective and desirable alternative to the use of venopuncture for the recovery of high yield genomic DNA for multiple assay applications.

The lower coefficient of variance of the Isohelix swab compared to that of swab A and swab B and the fact that the standard deviation is proportionally lower when compared to the group mean demonstrates that the high recovery from Isohelix swabs clearly shows that the high recovery of Nucleic acids form Isohelix swabs is very repeatable.

References

1. Qiagen Qiamp mini kit, part no 51504, Qiagen Ltd. Boundary Court, Gatwick Road, Crawley, West Sussex, RH10 2AX .

2. Standard protocol, Sciona Ltd, Method 001 rev 2.0, isolation of Genomic DNA from buccal swabs using the Qiagen Qiamp DNA mini kit, Sciona Ltd, Broadmarsh business and innovation centre, Harts farm way, Havant, Hampshire.

3. Picogreen dsDNA quantification reagent, part no P-7589, Molecular Probes Europe BV, PoortGebouw, Rijnsburgerweg 10, 2333 AA Leiden, The Netherlands.

4. Standard protocol, Sciona Ltd Method 004 rev2.01, Quantification of genomic DNA using Picogreen quantification reagent, Sciona Ltd, Broadmarsh business and innovation centre, Harts farm way, Havant, Hampshire.

5. Taqman assay, Applied Biosystems, Division Headquarters, 850 Lincoln Centre Drive, Foster City, CA 94404, U.S.A.