1. Introduction

Deciphering genomes/proteomes will not only help in researching hereditary predisposing factors but also contribute to designing drugs with higher efficacies. However, to arrive at this level of understanding we need to collect data in communities locally and globally. One of the major problems we face to achieve these objectives is volunteer recruitment for sample collection. Non-invasive sample collection represents the best option over blood in many ways:

- Better volunteer compliance
- No cold chain involvement for transport and storage
- Allows self-collection and does not require expertise

However, current non-invasive collection methods suffer from insufficient DNA yields, low quality, and impractical usability for the different segments of the population.

Invasive vs. Non Invasive Sampling

Invasive Sample Collection (Blood)
- Painful
- Requires expertise
- Expensive handling infrastructure (cold chain transport and storage)
- Low compliance

Non invasive collection technology alternative to blood that is compatible with simple and complex genomics downstream applications

2. Objective

- Provide a non-invasive sample collection technology that is compatible with simple and complex genomics downstream applications
- The iSWAB Sample Collection Technology
  - The squeezing insert allows the full release and concentration of cells from multiple swabs per patient
  - Unique design eliminates the need for swab transport
  - Immediate stabilization of collected sample at the point of collection

3. Methods

- **Sample collection:**
  1. Invasive: Drawing blood by a trained phlebotomist

- **The iSWAB Sample Collection Process**

- **DNA Extraction:**
  - Genomic DNA extraction was performed using QIAamp blood extraction kit.

- **DNA quantification:**
  - DNA yields where confirmed with 3 different methods: Nanodrop, picogreen assay and primers specific for the human beta actin gene were used to relatively quantitative the levels of human genomic DNA.

- **Bacterial DNA quantification:**
  - Bacterial DNA content was determined by using primers specific for the 65 ribosomal RNA gene.

- **PCR and Next Generation Sequencing (NGS):**
  - Direct PCR: 0.5 µL from iSWAB collected sample was directly added, without extraction, to a standard PCR master mixture. The samples where amplified using an MJ Research Thermal cycler.

- **NGS:**
  - Purified gDNA from both blood and iSWAB was used for the library generation and processing on MiSeq from Illumina.

4. Results

High Collection Compliance

These numbers were generated based on the response of 100 volunteers in reporting to designated locations to submit invasive and non-invasive samples.

High Genomic DNA Yield

DNA Yield from Various Age Groups per iSWAB

Age groups over 14 years old performed self collection. Samples from children under 14 were collected by a parent. Sample collection was performed at various times throughout the day.

5. Summary and Conclusions

- The high compliance and ease of use of iSWAB for all population segments including infants, toddlers, and elderly allows for achieving a better research outcome towards the understanding of genetic risk factors across various generations within families.
- iSWAB gDNA quality is comparable to blood allowing for field based collection and reducing energy costs from cold chain involvement.
- Proprietary tube insert design and lysing buffer yields up to 30µg of double-stranded, long fragment DNA ideal for simple and complex downstream genomic applications.
- Less than 1% bacterial genomic DNA contamination results in pure sample DNA leading for achieving more usable data.
- Swab-free sample transport reduces sample processing time without compromising sample integrity, resulting in faster turn around time.
- Long term room temperature stability reduces sample storage and transport costs by eliminating cold chain requirements.
- iSWAB implementation reduces operational cost significantly ($100Ks to $1000Ks)

Equivalent Performance to Blood gDNA in Illumina’s Targeted NGS Libraries

The proportional increase of PCR sensitivity to the gDNA input from iSWAB-DNA device is an indication of very limited PCR inhibitor carryover.

iSWAB-DNA Collected Samples are Stable Over 5 Years at Room Temperature Storage

iSWAB Preservation Capability Maintains the Integrity of gDNA at a Wide Range of Temperatures (-80°C to 65°C)

Storage Temperature

Low Bacterial Contamination (<1%)

Compared to Blood genomic DNA, the proportion of bacterial DNA contamination is significantly lower in iSWAB collected DNA.

No PCR Inhibition

Genomic DNA isolated from iSWAB-DNA was used as template to amplify a 300bp region of human genomic DNA (beta actin). 4% Agarose gel. 5 µl loaded. M: 25bp DNA marker, NC: Non-template control, lanes 1-24 replicates of the iSWAB sample.

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