

Invasive vs. Non-invasive Sample Collection: A look at DNA quality and its effect on downstream applications

Collection of whole blood is a well established source of high molecular weight of genomic DNA in abundant concentrations. Despite its widespread use, there are several drawbacks to using this technique:

- Painful
- Requires expertise
- Expensive and cumbersome handling infrastructure involving elaborate cold chain transport and storage

Successful genomic downstream applications are highly dependent on the quality of extracted gDNA. Current blood collection vessels containing-coagulants such as residual heme are well known to be PCR inhibitors.

Non-invasive collection methods represent a viable alternative to blood collection for several reasons:

- Painless
- Enables self or assisted collection
- No cold chain involvement

However, traditional non-invasive collection methods suffer from high level of bacterial contamination, low gDNA yield, or highly fragmented DNA.

The iSWAB-DNA non-invasive collection device represents a novel solution that offers all the advantages of non-invasive collection with almost none of the drawbacks. iSWAB technology utilizes a squeezing mechanism that allows concentration of oral cells from multiple samples. The concentrated cells are gently lysed and content is stabilized in the iSWAB proprietary lysis buffer. This unique collection mechanism allows for high quality long fragment ds DNA at high concentration that contains <1% bacterial DNA.

The effect of the source of gDNA and its input quantity on the generation of Illumina's targeted sequencing libraries PCR sensitivity

C5/C7 in the following figures refers to the Illumina library adapters used to amplify targeted sequencing libraries utilizing gDNA extracted from blood, saliva and the iSWAB collected samples. The gel images shown are library products that came from a PCR performed at the end of AbVitro's proprietary targeted sequencing method.

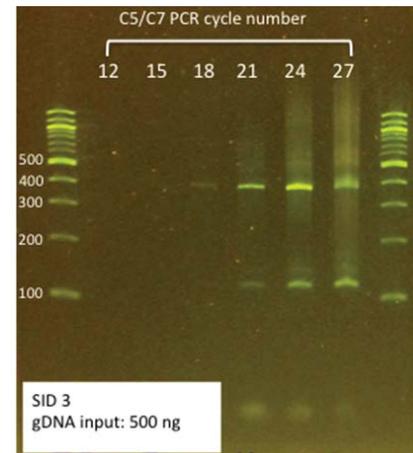


Fig 1. C5/C7 PCR cycle numbers from gDNA purified from iSWAB-DNA collection device, 500 ng

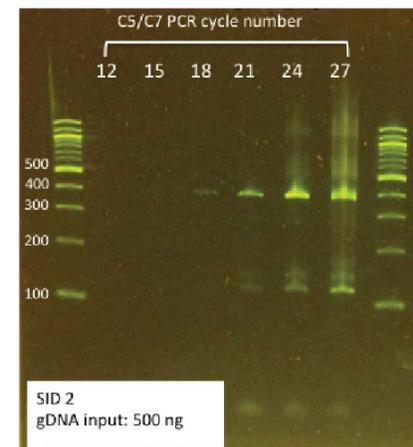


Fig 2. C5/C7 PCR cycle numbers from gDNA purified from EDTA-Blood, 500 ng

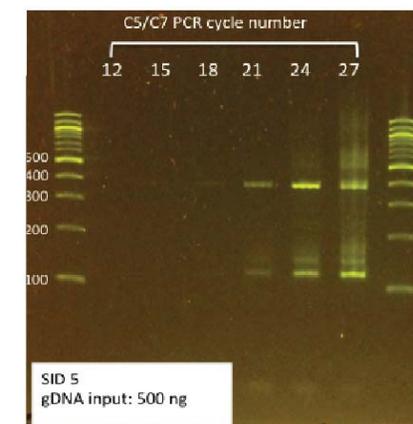


Fig 3. C5/C7 PCR cycle numbers from gDNA purified from saliva collection device, 500 ng

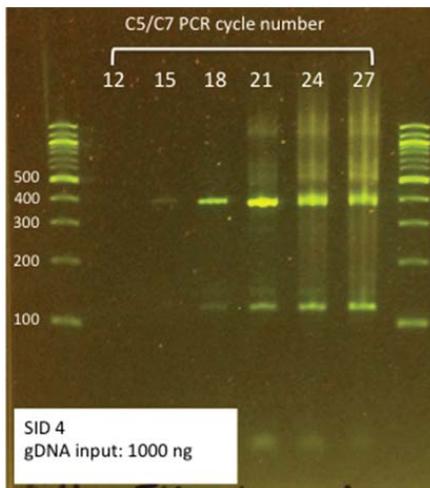


Fig 4. C5/C7 PCR cycle numbers from gDNA purified from iSWAB-DNA collection device, 1000 ng

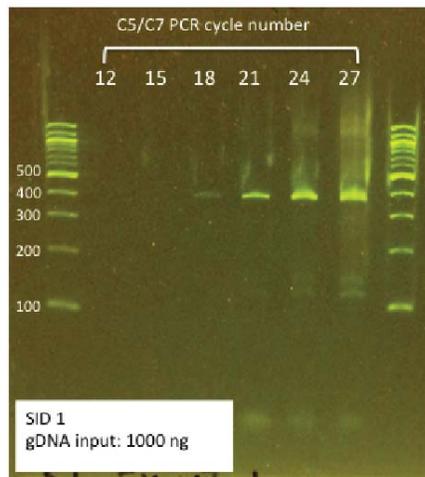


Fig 5. C5/C7 PCR cycle numbers from gDNA purified from EDTA-Blood, 1000 ng

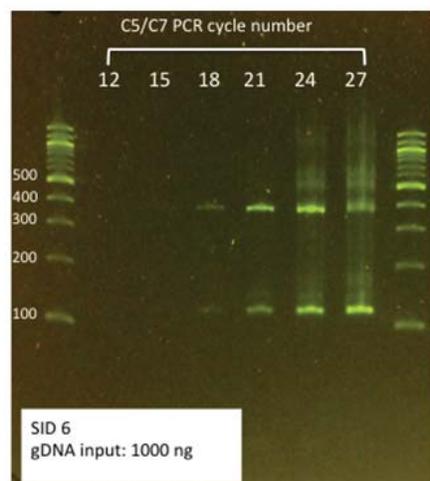


Fig 6. C5/C7 PCR cycle numbers from gDNA purified from saliva collection device, 1000 ng

Results

At 500ng gDNA input, there was insignificant difference observed in PCR sensitivity for gDNA extracted from the three different sample types (Fig 1 to 3). However, at higher gDNA input (1000ng), the PCR sensitivity decreased significantly; which is a phenomenon usually observed in association with increased PCR inhibitors at higher amount gDNA inputs. This effect has not been observed with the gDNA input extracted from iSWAB-DNA collection device (Fig 4 to 6). Products from both libraries, C5/C7 were detected at the 15th PCR cycle when gDNA purified from the iSWAB samples were used, however blood and saliva lagged. **The proportional increase of PCR sensitivity to the gDNA input from ISWAB-DNA device is an indication of very limited PCR inhibitor carryover.**

Summary and Conclusions

The iSWAB-DNA performed as well or better than blood and saliva collection methods and enabled increased PCR detection sensitivity.

The iSWAB-DNA is an ideal non-invasive oral sample collection method and compatible with simple and complex genomics downstream applications.

**Data generated in this application note is a courtesy of AbVITRO, Inc.*



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