One-hour automated HMW DNA extraction from blood and saliva for HiFi sequencing

Deborah Moine, Shreyasee Chakraborty, Christine Lambert, Harsharan Dhillon, Ian McLaughlin, Juniper Lake, Jeff Zhou, Davy Lee and Heather Ferrao
PacBio, 1305 O'Brien Drive, Menlo Park, CA 94025

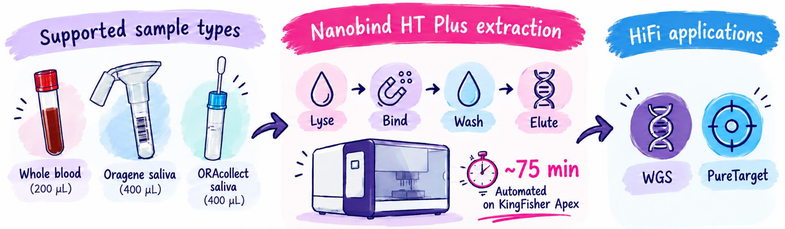
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Automated HMW DNA extraction from blood and saliva

Background

- Blood and saliva are important sample types for large-scale genomic studies. Saliva offers the advantages of non-invasive collection and room-temperature stability.
- HiFi sequencing requires high-quality, high-molecular-weight (HMW) DNA for optimal long-read sequencing performance.
- We developed the Nanobind HT Plus kit, enabling rapid, automated HMW DNA extraction from whole blood, and saliva collected in Oragene (TM) and ORAcollect devices.

Workflow overview



Extraction performance

308 samples were extracted using the Nanobind HT Plus kit on the KingFisher Apex system: 36 ORAcollect saliva, 108 Oragene saliva, and 164 whole blood samples.

- High-quality HMW DNA was recovered across all supported sample types: mean DNA yield of 5.9 µg for ORAcollect saliva, 12.5 µg for Oragene saliva, and 9.4 µg for whole blood (200 µL).
- ORAcollect saliva showed lower DNA yield and GQN10 kb, consistent with its lower-input collection format.
- Yield variability reflects biological differences in cellular/WBC content across donors and collection formats.

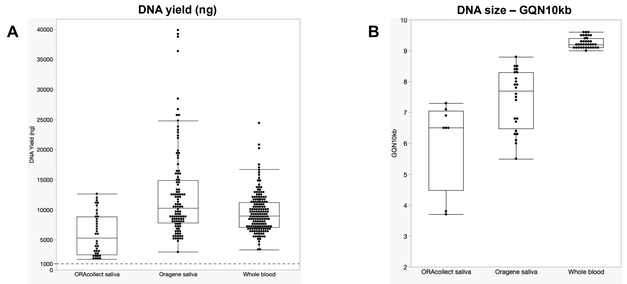


Figure 1. Extraction yield and HMW DNA quality across supported sample types.
DNA yield **(A)** and GQN10kb **(B)** for ORAcollect saliva (swab saliva), Oragene saliva, and whole blood. Dashed line in **(A)**: 1000 ng DNA yield threshold.

HiFi WGS performance

HiFi WGS libraries were prepared from Nanobind HT Plus–extracted DNA using standard PacBio library preparation procedures and sequenced on the Revio system using SPRQ-Nx reagents.

- 56 samples across 28 Revio acquisitions (2 samples multiplexed per aquisition): 4 ORAcollect saliva, 12 Oragene saliva, and 12 whole blood.
- Robust HiFi yield across all supported sample types: mean HiFi yield of 128 Gb for ORAcollect saliva, 139 Gb for Oragene saliva, and 142 Gb for whole blood.
- ORAcollect saliva shows lower HiFi yield and read length, consistent with its lower-input collection format.

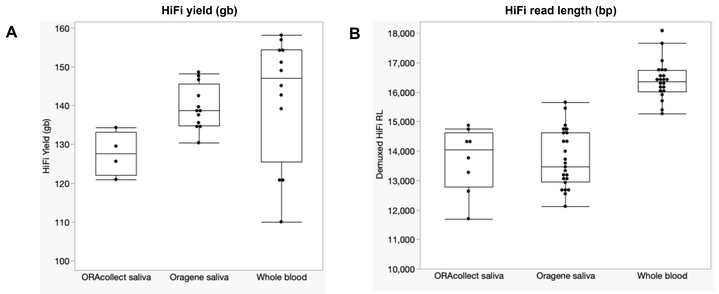


Figure 2. HiFi WGS performance across supported sample types. HiFi yield **(A)** and read length **(B)** for ORAcollect saliva (swab), Oragene saliva, and whole blood extracted with the Nanobind HT Plus workflow..

Human read mapping

HiFi reads were mapped to the human reference genome (GRCh38) using SMRT Link to determine the percentage of human-mapped reads.

High human mapping rates (88.5–98.0%) were observed across the analyzed Oragene saliva and ORAcollect saliva samples.

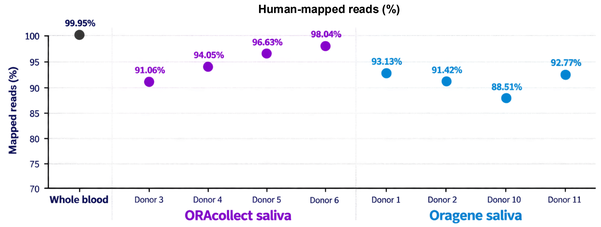


Figure 3. Human read mapping across supported sample types. Percentage of HiFi reads mapped to GRCh38 for ORAcollect saliva (n=4 donors) and Oragene saliva (n=4 donors) following automated Nanobind HT Plus extraction. Whole blood (99.95%) is shown as a reference.

PureTarget performance

PureTarget 24 Carrier panel libraries were prepared from Oragene saliva DNA extracted using the Nanobind HT Plus kit and sequenced on the Revio system.

- 12 donors run in duplicate (24 samples)
- Mean target coverage: 234X (range: 100–400X)
- All Oragene saliva samples met the ≥20x target coverage minimum and the ≥40x minimum for gene/pseudogene targets

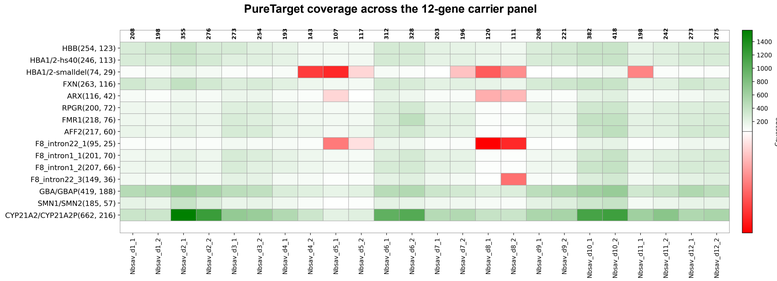
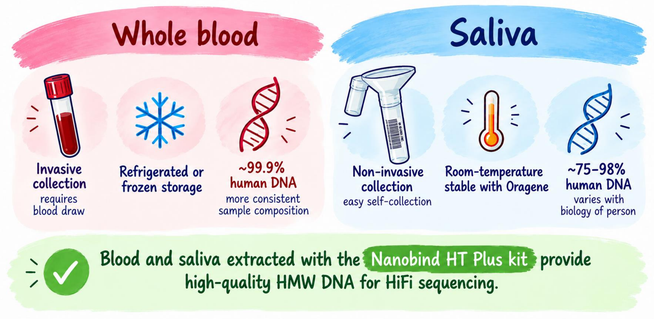


Figure 4. PureTarget carrier-panel coverage from Oragene saliva. Twelve donors were analyzed in duplicate following automated Nanobind HT Plus extraction. All 24 samples met the required target coverage thresholds. Values in parentheses indicate the mean and minimum coverage for each target across all samples, while values above the heatmap indicate the mean coverage across all targets for each sample. Colors represent target coverage.

Conclusion



Acknowledgements

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