

Targeted HiFi sequencing at scale with ready-made Twist Alliance Panels or custom designs

Combine [Twist Bioscience](#) target enrichment with [PacBio](#) long and accurate HiFi reads to efficiently sequence your priority genomic regions at scale. Sequence enriched regions with a protocol optimized for HiFi reads to get comprehensive detection of single nucleotide variants, structural variants, and indels with haplotype resolution. HiFi target enrichment can deliver accurate alleles for complex gene families such as immune genes (e.g., *HLA*) and pharmacogenes (e.g., *CYP2D6*).



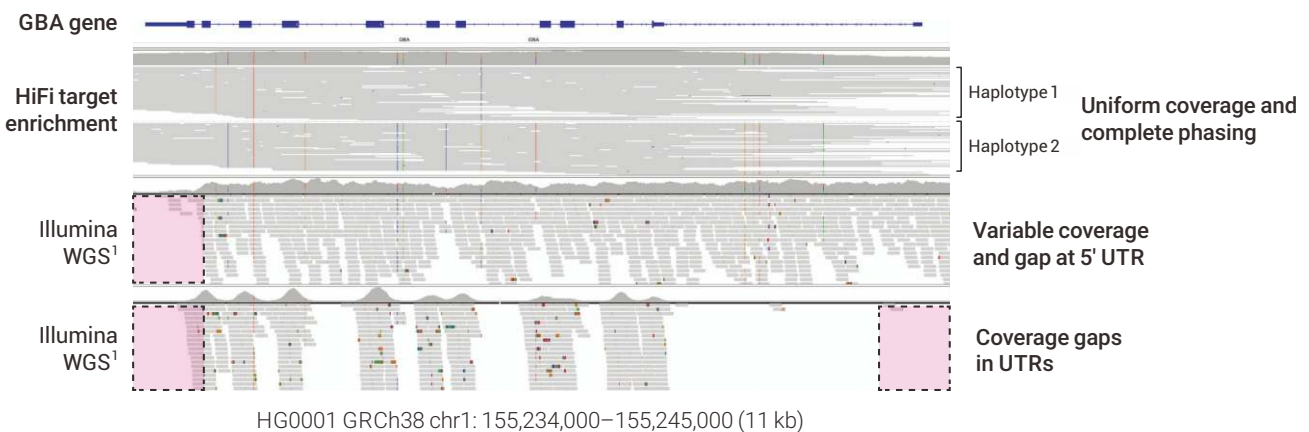
Why choose HiFi sequencing with Twist target enrichment?

- End-to-end workflow optimized for HiFi reads
- Custom design for any panel size
- Small and large cohorts on a single SMRT[®] Cell
- High-accuracy variant calls, including SVs
- Direct phasing, unambiguous haplotypes, ancestr-agnostic discover

Pool multiple samples per SMRT Cell

| Panel example | Large panel | Medium panel | Small panel |
|--|-------------|--------------|-------------|
| Panel size | 20 Mb | 2 Mb | 100 kb |
| Estimated number of genes | 400 | 50 | 2 |
| Samples per Sequel [®] IIe SMRT Cell 8M | 4 | 24 | 96 |
| Samples per Revio [™] SMRT Cell | 12 | 72 | 288 |

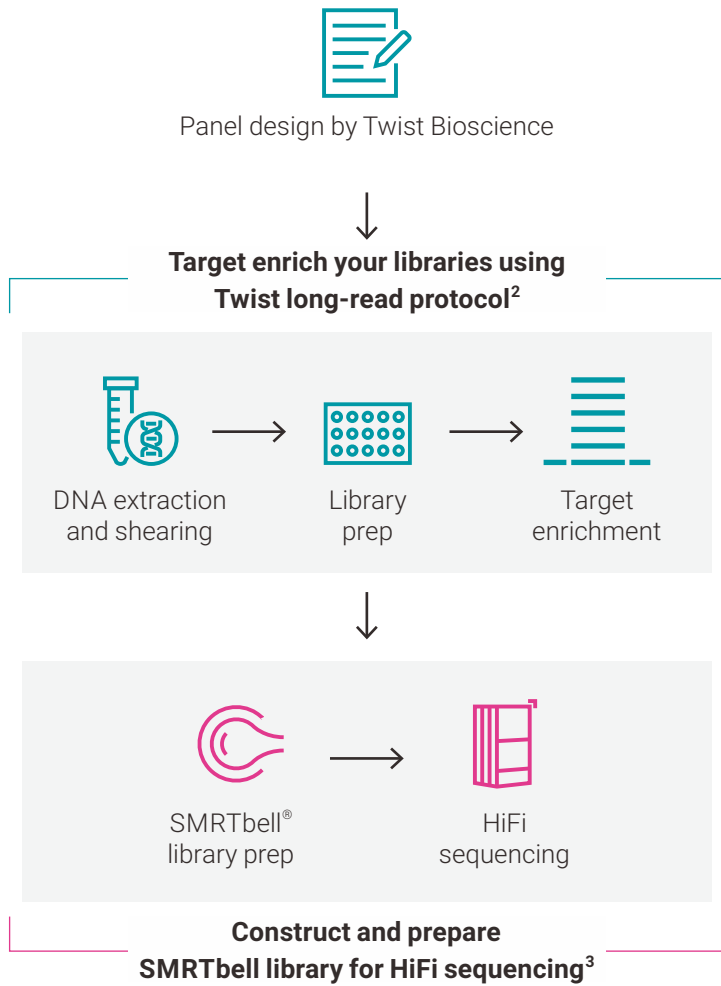
Full gene coverage of medically relevant genes



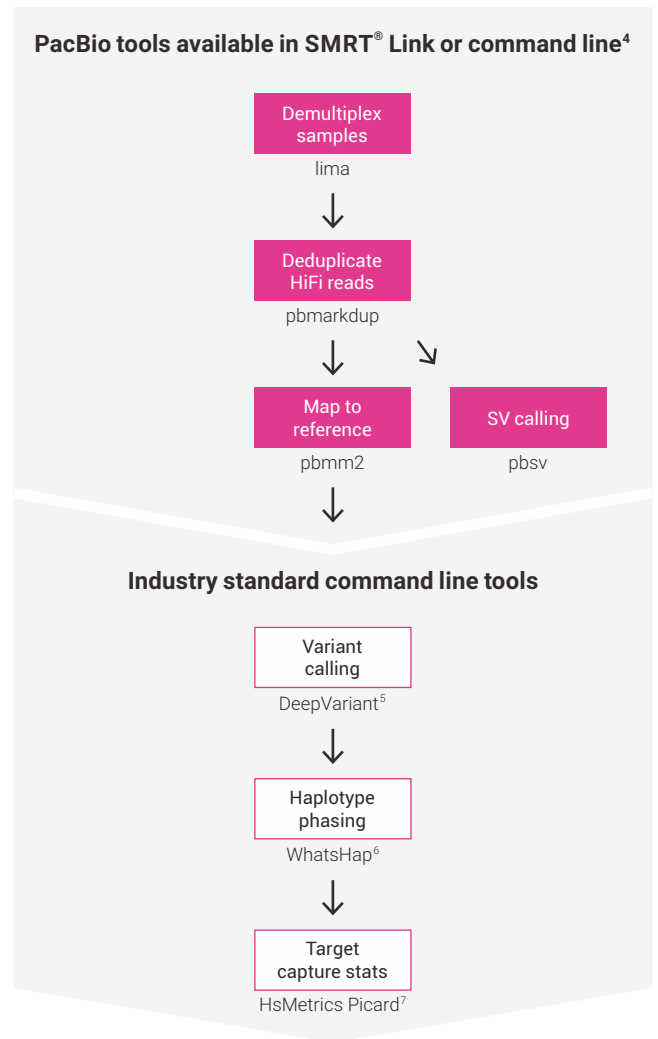
Performance for Twist Alliance Panels

| Twist Alliance Panel | Panel Size (Mb) | Number of genes | PacBio system | Samples per SMRT Cell | Mean target coverage | Fold enrichment | Target bases ≥ 10 -fold read depth | Mean HiFi read length | Duplicate rate |
|----------------------|-----------------|-----------------|---------------|-----------------------|----------------------|-----------------|---|-----------------------|----------------|
| Dark Genes | 22 | 389 | Revio | 12 | 75-fold | 65-fold | 93% | 5.5 kb | 3% |
| Dark Genes | 22 | 389 | Sequel IIe | 4 | 75-fold | 54-fold | 93% | 5.2 kb | 3% |
| Long Read PGx | 2 | 49 | Sequel IIe | 24 | 190-fold | 784-fold | 99% | 5.3 kb | 2% |

From targets to HiFi reads



Recommended analysis tools



REQUIRED REAGENTS

| Vendor | Quantity | Description | PN | Quantity supported |
|-----------------------|----------|---|-------------|-----------------------|
| Twist Bioscience | 1 | Twist Library Prep MF Kit 1 & 2 | 101281 | 96 samples |
| | 1 | Twist Universal Adapter System Plate A | 101308 | 96 samples |
| | 2 | Twist Library Prep Kit 2 (AMPure beads) | 100573 | 96 samples |
| | 1 | Twist Universal Blockers, 12 reactions | 100578 | 12 captures |
| | 1 | Twist Standard Hyb and Wash Kit v2 | 104446 | 12 captures |
| | 1 | Custom Probe Panel, 12 reactions | 101001 | 12 captures |
| PacBio | 1 | SMRTbell prep kit 3.0 | 102-182-700 | 24 SMRTbell libraries |
| | 1 | Binding kit 3.2 and cleanup beads | 102-333-300 | 24 binding reactions |
| | 1 | SMRT Cell 8M tray | 101-389-001 | 4 SMRT Cells |
| | 1 | Sequel [®] II sequencing kit 2.0 | 101-820-200 | 4 SMRT Cells |
| Invitrogen | 1 | Dynabeads M-270 (2 mL vol) | 65305 | 20 captures |
| Merck/Millipore/Sigma | 1 | KOD Hot Start DNA Polymerase (200 units) | 71975 | 44 samples |

KEY REFERENCES

- <https://www.biorxiv.org/content/10.1101/2020.12.11.422022v1.full> (HG002.novaseq.wes-agilent.50X)
- <https://www.twistbioscience.com/resources/protocol/long-read-library-preparation-and-standard-hyb-v2-enrichment>
- [Procedure & checklist – Preparing multiplexed amplicon libraries using SMRTbell prep kit 3.0](#) PacBio documentation.
- <https://github.com/PacificBiosciences/>
- <https://github.com/google/deepvariant>
- <https://whatshap.readthedocs.io/en/latest/>
- <https://snakemake-wrappers.readthedocs.io/en/stable/wrappers/picard/collecthsmetrics.html>

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