Guide to high-molecular-weight DNA extraction and long-read sequencing

Looking for the right high-molecular-weight genomic DNA kit for long-read sequencing?

Long-read sequencing using PacBio® and Oxford Nanopore Technologies® generate extensive reads for complex genomic studies but it is sensitive to DNA quality and integrity, requiring high-quality, contaminant-free genomic DNA (gDNA). Sample preparation challenges include avoiding DNA contamination, degradation, or shearing to ensure accurate sequencing results. To address these, we offer sample preparation kits for high-molecular-weight (HMW) gDNA samples designed to reduce costs and processing times while maintaining DNA quality and integrity.

Follow these 3 steps to find the right kit for you:

1 What's the length of the genomic DNA?

2 What's your sample type and input amount?

Typical gDNA Size	Soil	Stool	Bacteria	Yeast	Cells	Tiss
					up to 5×10^7	200
~100 kb						
			$2.2 \times 10^{10} - 1 \times 10^{11}$	$7 \times 10^9 - 3.5 \times 10^{10}$	$2 \times 10^7 - 1 \times 10^8$	100–4
50–100 kb			$2.2 \times 10^{10} - 1 \times 10^{11}$	$7 \times 10^9 - 3.5 \times 10^{10}$	$2 \times 10^7 - 1 \times 10^8$	100–4
			2×10^9			up to
					5×10^6	up to
10–20 kb					up to 5×10^6	up to
		up to 250 mg				
	up to 250 mg					
	up to 10 g					

Scan this QR code to learn more about our HMW gDNA extraction kits
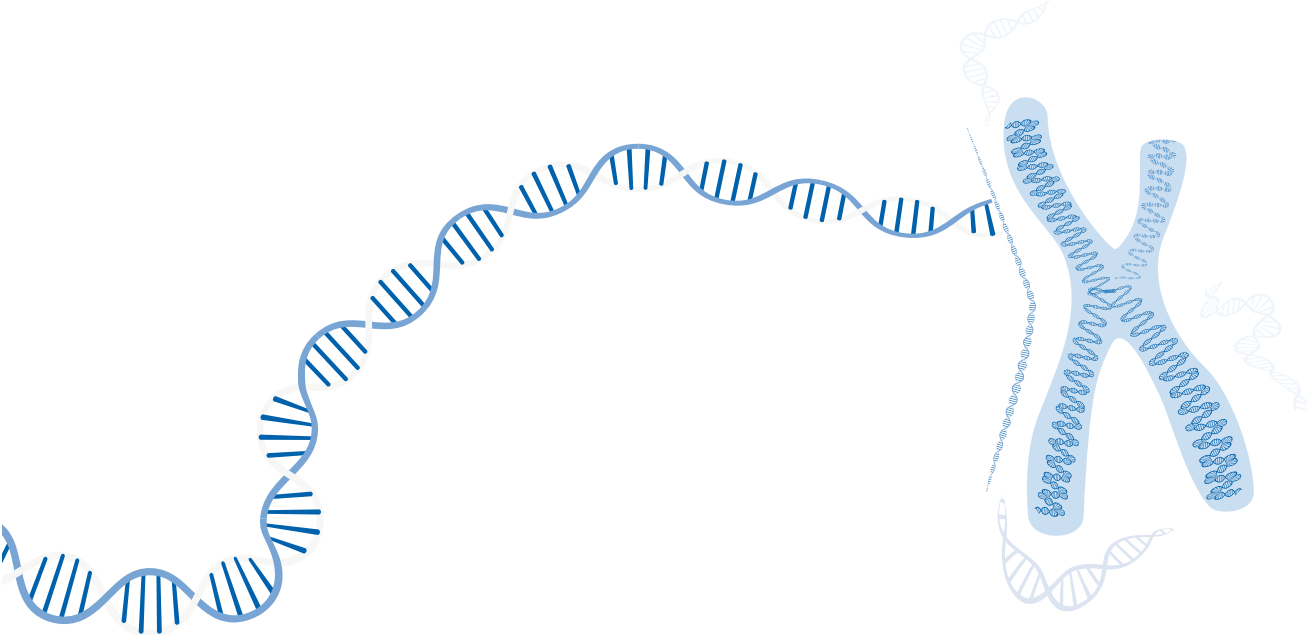
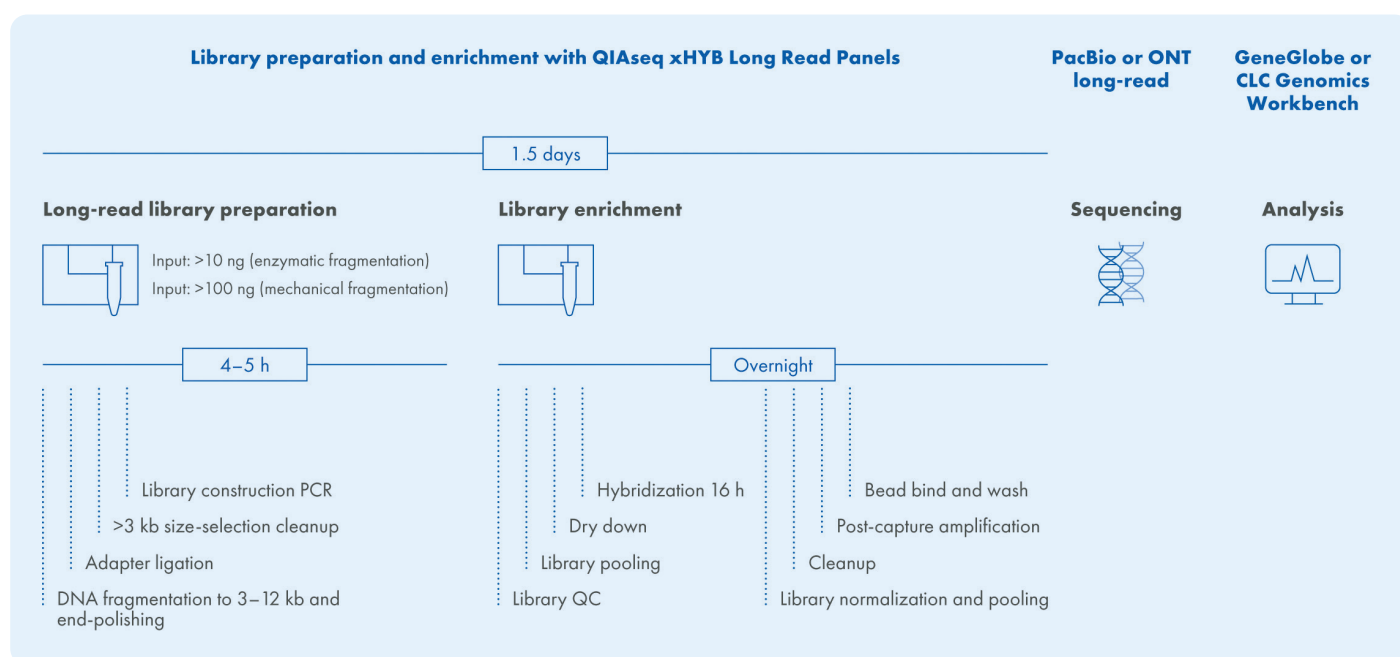| ue	Blood		Method		Product
0 mg	200 µL – 10 mL		Precipitation		**Puregene® Kits**
	8.5 mL		Precipitation		**PAXgene® Blood DNA Kit**
400 mg	1–20 mL		Anion exchange		**QIAGEN® Genomic-tips (only Midi & Maxi)**
400 mg	1–20 mL		Anion exchange		**QIAGEN Blood & Cell Culture DNA Kit**
25 mg	200 µL		Beads		**MagAttract® HMW DNA Kit**
	200 µL		Spin column		**QIAamp® DNA Blood Mini Kit**
25 mg			Spin column		**QIAamp DNA Mini Kit**
25 mg	100 µL		Spin column		**DNeasy® Blood & Tissue Kits**
			Spin column		**QIAamp PowerFecal® Pro DNA Kit**
			Spin column		**DNeasy PowerSoil® Pro Kit**
			Spin column		**DNeasy PowerMax® Soil Kit**

Play the long game in genomics powered by long reads

With high molecular weight DNA in hand, it's crucial to maintain that integrity throughout the library preparation process to fully realize the benefits of long-read sequencing technologies.

QIAseq® xHYB Long Read Panels deliver the resolution you need to tackle the most complex genomic regions using the power of long-read sequencing.

- Complete, uniform coverage with hybrid capture-based target enrichment
- High sensitivity and specificity for accurate variant detection
- Compatible with long-read sequencing for accurate variant calling and phasing
- Flexible and customizable to match diverse research needs and budgets
- Compatible with PacBio (Revio and Vega long-read systems) and ONT long-read systems

Long-read NGS library preparation and target enrichment workflow

Library preparation and enrichment with QIAseq xHYB Long Read Panels   **PacBio or ONT long-read**   **GeneGlobe or CLC Genomics Workbench**

1.5 days

Long-read library preparation   **Library enrichment**   **Sequencing**   **Analysis**

Input: >10 ng (enzymatic fragmentation)
Input: >100 ng (mechanical fragmentation)

4–5 h   Overnight

Library construction PCR   Hybridization 16 h   Bead bind and wash
>3 kb size-selection cleanup   Dry down   Post-capture amplification
Adapter ligation   Library pooling   Cleanup
DNA fragmentation to 3–12 kb and end-polishing   Library QC   Library normalization and pooling

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HLA Typing Panel

Comprehensive panel design covers all 18 HLA class I and II genes, including introns and UTRs, enabling high-resolution 4-field haplotyping

Hereditary Cancer Panel

Targets over 1000 gross deletions, duplications, insertions and complex rearrangements linked to a wide range of cancer types

Custom Panel

Fully customizable panel design targets your specific regions of interest, enables integration with existing QIAseq xHYB long-read or custom panels and boosts coverage with additional probes – no ROI limits
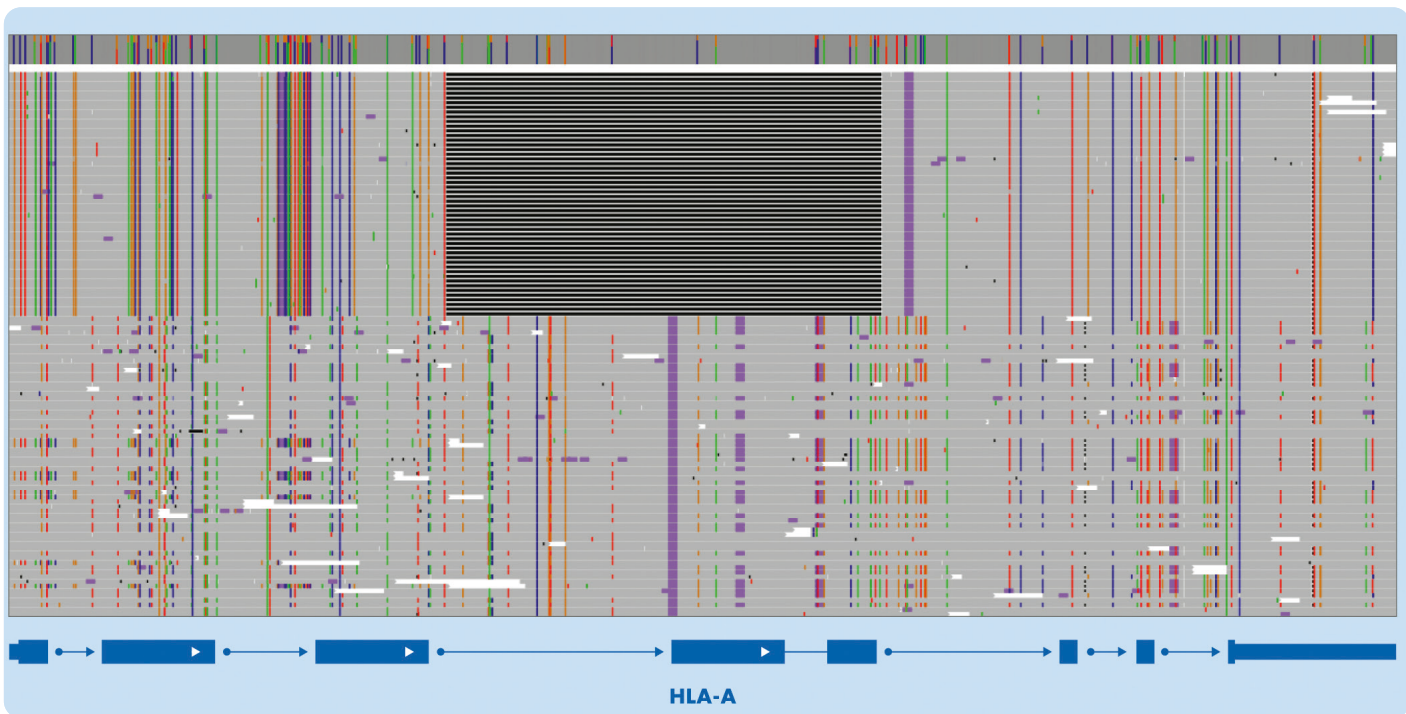
Figure 1: Long reads generated using the QIAseq xHYB Long Read HLA Typing Panel span the HLA-A gene without assembly, simplifying haplotyping. Here, we demonstrate the long reads that span the full length of the HLA-A gene, eliminating the need for complex assembly and simplifying the haplotyping process. Visualized read stacks across the gene clearly show heterozygous polymorphisms, with colored markers indicating roughly equal representation of non-reference bases – consistent with the gene's natural allelic diversity.
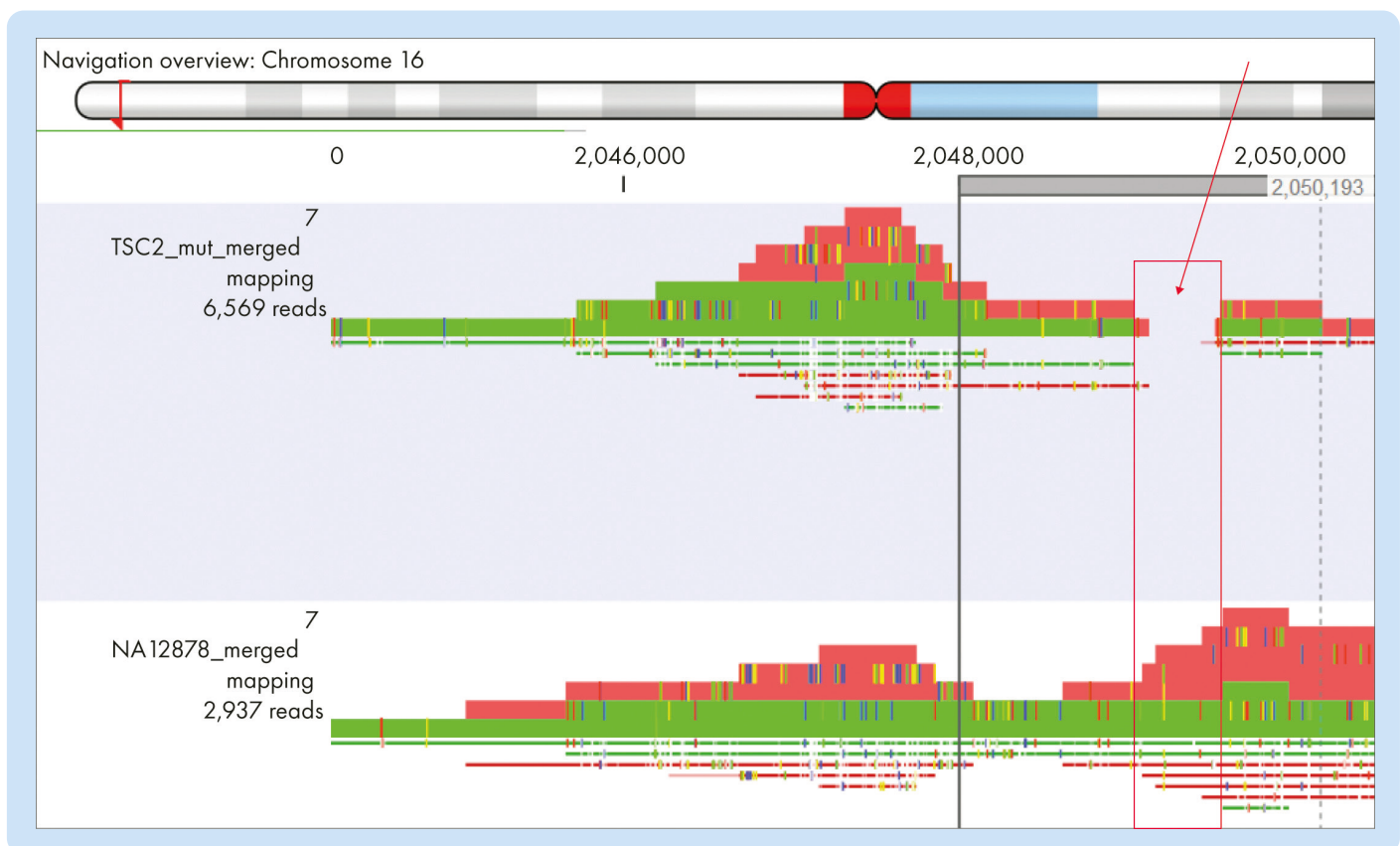

Figure 2: QIAseq xHYB Long Read Hereditary Cancer Panel successfully detects TSC2 deletion after exon 1 through long reads and contiguous mapping. The QIAseq xHYB Long Read Hereditary Cancer Panel accurately identifies a large TSC2 deletion in sample NA04520, a structural variant often challenging for short-read methods. The image displays long-read mapping data generated using CLC Genomics Workbench, comparing Coriell reference DNA NA04520 from an affected individual with benchmark reference DNA NA12878. The zoomed-in view of the 5' end of the TSC2 gene clearly shows a 430 bp deletion in NA04520 (indicated by the red arrow and coverage gap).

Applications

Repeat expansion disorder analysis
Precisely characterize repeat expansions in neurological and genetic disorders

Phasing of genetic variants
Determine haplotype structures of genomic variants for critical insights into inheritance patterns and allele-specific expression

HLA typing
Resolve polymorphic HLA regions for accurate genotyping in immunogenomics and transplantation

Structural variant detection
Accurately detect large-scale genomic alterations for cancer, genetic disease and evolutionary research

Data analysis

The GeneGlobe® Data Analysis portal offers simplified downstream data analysis for HLA haplotyping. The QIAGEN® CLC Genomics Workbench is used to detect large structural variants in hereditary cancers.

To learn more about the long-read sequencing panel, visit **https://www.qiagen.com/qiaseq-long-read-seq**

Visit our **Genomic DNA Resource Center for High-Molecular-Weight DNA** and advance your exploration of long-read sequencing.

Ordering	**www.qiagen.com/shop**
Technical Support	**www.support.qiagen.com**
Website	**www.qiagen.com**