xGen™ HIV Amplicon Panel

Comprehensive coverage to elevate your HIV research

Save critical time

Trace variants with confidence

Stocked and ready to ship

IDT's xGen NGS solutions are ready to advance the science community in its quest to monitor the evolution of HIV.

Our uniquely configured, single-panel design creates overlapping amplicons to reliably distinguish identified and unidentified viral variants. Our differentiated super amplicon technology overcomes amplicon drop out and increases coverage for variable genomes, which is essential for tracing evolving epidemiological patterns.

- **Increase your coverage**—achieve >99% coverage of the HIV genome
- **Super amplicons**—coverage possible even if a primer dropout occurs
- **Work faster, not harder**—workflow is <2.5 hr for library prep with <1 hr hands-on time
- **Normalize multiplexed samples easily**—optional Normalase™ technology provided
- **Automate your work**—up to 1536 UDIs for comprehensive data analysis
- **Customize it**—spike-in more primers to make the panel work for you

Table 1. Features of the xGen HIV Amplicon Panel.

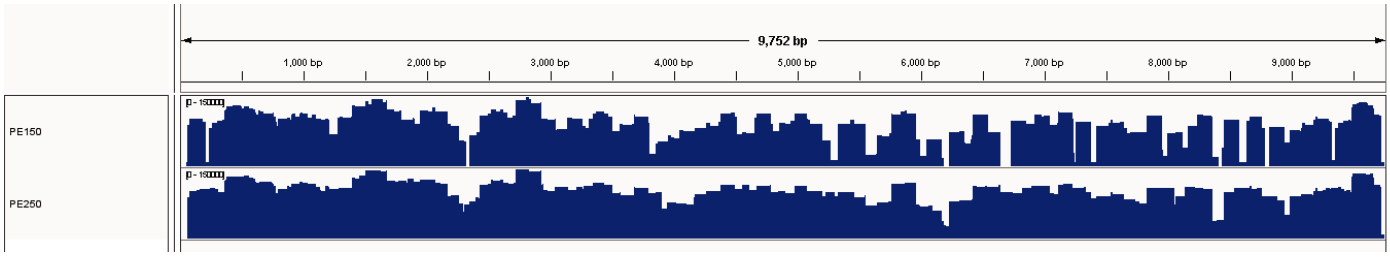
Features	Specifications
Design coverage and panel information	107 amplicons, average amplicon size 102 bp Primers designed to target HIV (HXB2, GenBank K03455) Minimum 1,000 double stranded viral genome copies
Input material	1st or 2nd strand cDNA, dsDNA 1,000 dsDNA viral copies
Time	2.5 hours viral DNA-to-library
Multiplexing capability for Illumina	Up to 1536 UDIs
Compatible with other indexes for Illumina?	Yes
Recommended depth for Illumina	500,000 reads per library, PE250 or PE150

Sequence the entire HIV genome

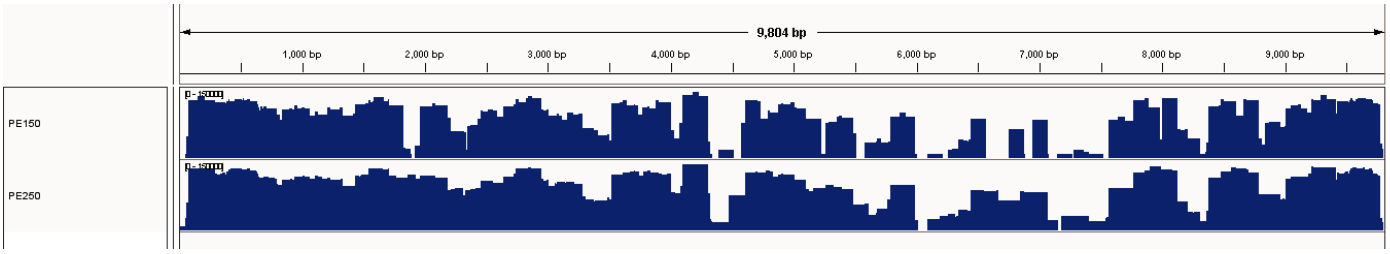
The xGen HIV Amplicon Panel offers a streamlined (cDNA-to-sequencer in 2.5 hours), single-tube NGS workflow for studying the entire Human Immunodeficiency Virus genome, not just specific genes (**Figure 1**). This Predesigned xGen Amplicon Panel built with xGen Amplicon Technology includes amplicon tiling and creation of super amplicons to ensure comprehensive genome coverage across existing variant genomes and provide resistance to future viral mutations that may fall within a priming site, thus enabling future identification of novel variants (see our Tech Note on how **xGen Amplicon panels leverage super amplicon technology).**

Due to super amplicon formation, a high level of coverage across the complete variant genomes can be preserved (**Figure 1**). The xGen HIV Amplicon Panel demonstrates high percent mapping and on-target reads to the HIV genome (**Table 2**).

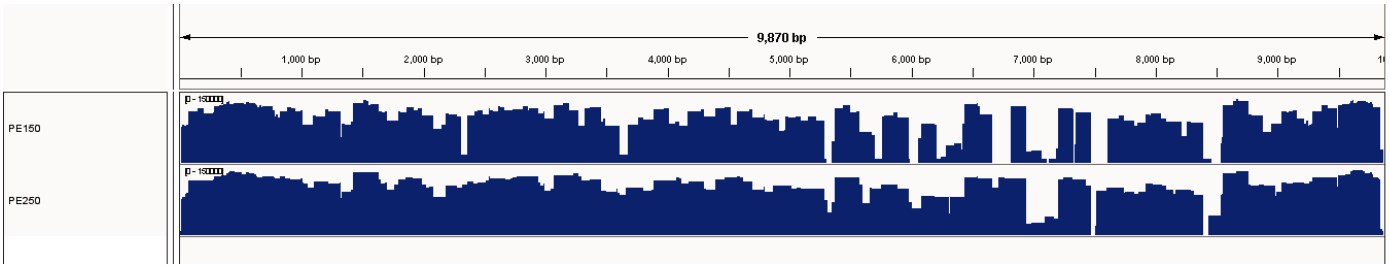
A. HIV plasmid 409 genome coverage



B. HIV plasmid 421 genome coverage



C. HIV plasmid 439 genome coverage



D. HIV plasmid 454 genome coverage

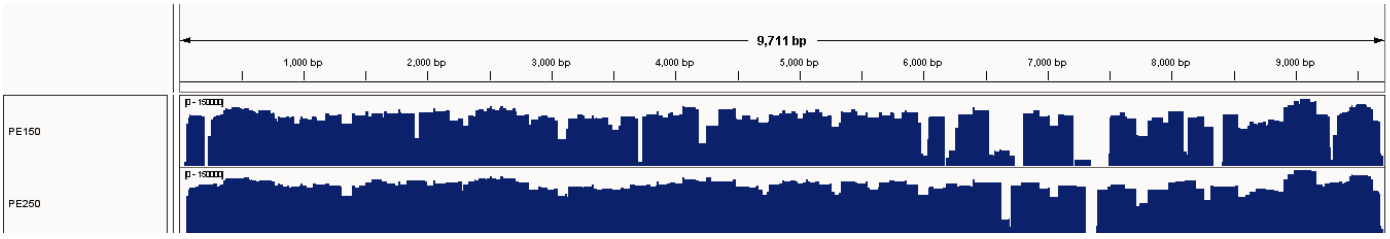


Figure 1. The xGen HIV Amplicon Panel provides a high level of coverage across HIV variant genomes. Examples of panel coverage are shown for four variants. For all libraries, 1,000 plasmid copies carrying the complete HIV genome from four different strains ranging from 91–93% identity to the HXB2 reference genome were tested individually (plasmids described in Zaikos et al., 2018) in a background of 10 ng human gDNA (Coriell NA12878). The resulting NGS libraries generated with the xGen HIV Amplicon Panel were sequenced on a MiSeq™ (Illumina) with 150 bp and 250 bp paired-end sequencing (PE150 top coverage plot, PE250 bottom coverage plot). Resulting reads were subsampled to 500,000 total reads per sample for analysis. Example coverage data for one replicate per HIV genome are shown in panels **A–D**. The reference genomes used as input are indicated above (**A–D**). Despite the variation in these genomes to the HXB2 panel design genome, coverage is maintained across each genome due to super amplicon formation. The increase in coverage observed with the PE250 read length is due to improved coverage of the super amplicons spanning highly variable regions, which are not fully accessible with the shorter PE150 read length.

Although the variant genomes had only 91–93% identity to the HXB2 reference genome used to design the panel, the formed super amplicons maintained coverage across multiple variants' genomes (**Table 2**).

Table 2. xGen HIV Amplicon Panel NGS metrics on Illumina.

	Plasmid 409		Plasmid 421		Plasmid 439		Plasmid 454	
	PE150	PE250	PE150	PE250	PE150	PE250	PE150	PE250
% identity to HXB2 design	92%		91%		93%		93%	
% mapping	93.8%	94%	95.7%	95.8%	92.4%	92.6%	95.7%	95.8%
% on-target (base)	94.5%	94.3%	81.7%	82.3%	82.7%	83.8%	94%	93.2%
% base uniformity	53.9%	66.6%	53.9%	57.5%	65.4%	76.4%	75.9%	87%
% genome >10x coverage	92.5%	99.3%	80.9%	91.5%	88.5%	98.6%	91.7%	98.5%
% genome covered	97%	99%	92%	98%	95%	99%	96%	100%

xGen amplicon sequencing technology—super amplicons cover the genome

xGen Amplicon Technology from IDT enables single-tube multiplexed PCR even when contiguous target coverage of overlapping amplicons is needed, unlike other methods that require multiple tubes for target enrichment. In this methodology, primers are designed to compensate for changes in the target genomic DNA. If one of the primers cannot anneal to a location due to a mutation, then neighboring primers can compensate for the loss, and amplify the region as a longer amplicon—a super amplicon—that provide you complete coverage of novel variants and identification of genotypic evolution.

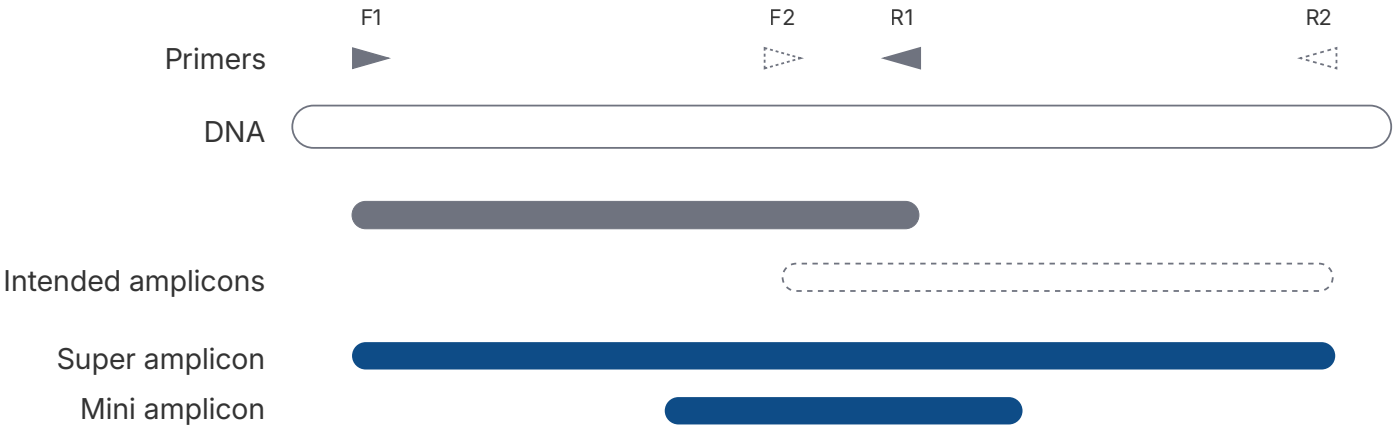


Figure 2. xGen Amplicon Sequencing Panels are a one-tube solution for coverage. With the xGen Amplicon sequencing single-tube workflow, there is high coverage even when an individual primer (such as primer F2) anneals less efficiently due to a genomic mutation in its binding site. Super amplicons are created when the nearby primer pair, F1 and R2, amplify the area including the variant.

References

1. Zaikos TD, Terry VH, Sebastian Kettinger NT, et al. **Hematopoietic Stem and Progenitor Cells Are a Distinct HIV Reservoir that Contributes to Persistent Viremia in Suppressed Patients**. Cell Rep. 2018;25(13):3759-3773.e9. doi:10.1016/j.celrep.2018.11.104

Ordering information

Product	Catalog #
xGen HIV Amplicon Panel 96 rxn	10020328
xGen Amplicon UDI primers Plate1	10009847
xGen Amplicon Core 96rxn	10009827

For more information, visit **idtdna.com/NGSHIV**