Product Profile

QIAseq FX DNA Library Kit

For high-performance, all-enzymatic whole genome and hybrid capture library preparation for Illumina® instruments

Since its introduction, next-generation sequencing (NGS) has found numerous applications with considerable impact and potential, including cancer research, stem cell research, metagenomics, population genetics and biomedical research. While NGS technology is continuously improving, library preparation remains one of the main bottlenecks as it involves several time-consuming steps. Not only can library preparation result in considerable sample loss, there is also the potential to introduce handling errors.

QIAGEN QIAseq FX technology incorporates all-enzymatic DNA fragmentation into a streamlined, optimized protocol that does not require sample cleanup between fragmentation and adapter ligation. This saves time and prevents errors. Optimized enzyme and buffer compositions ensure high sequencing library yield. A simple, three-reaction protocol enables straightforward automation of library preparation on various liquid-handling platforms, reducing hands-on time and run-to-run variability.

Benefits of the QIAseq FX DNA Library Kit:

- Sequencer-ready libraries from genomic DNA in just 2.5 hours
- Fragment size, input amount and batch size customizable to any sequencing experiment
- Low G/C bias for even genomic coverage
- Superior performance compared to other enzymatic fragmentation methods
- Convenient kit format including dual-barcoded adapters

Fast, convenient, all-enzymatic library preparation

Overcome the limitations of current short-read NGS DNA fragmentation technologies. The new QIAseq FX DNA Library Kit takes you from 1 ng – 1 µg genomic DNA to sequencer-ready, amplified libraries in just 2.5 hours (Figure 1). It's faster and easier to automate than mechanical shearing but still generates the high-quality libraries you need for whole genome sequencing of any organism.

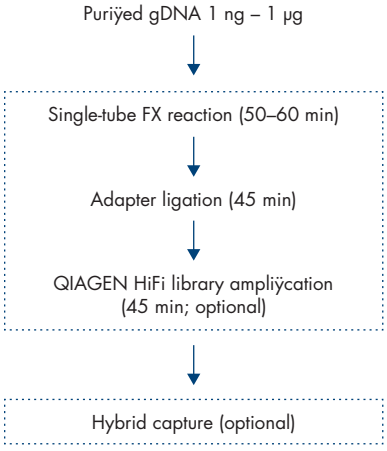

Figure 1. Streamlined three-step workflow. Go from purified gDNA to sequencer-ready libraries in just 2.5 hours with the QIAseq FX DNA Library Kit.

The simple workflow has 2 steps, plus an optional library amplification, and involves less than 20 minutes hands-on time, providing unmatched convenience (Figure 1). Starting with as little as 1 ng input DNA, the QIAseq FX chemistry fragments, end repairs and A-tails the DNA in under 1 hour. This is followed by high-efficiency adapter ligation and an optional, high-fidelity amplification step using the proprietary HiFi Master Mix. The workflow generates reliably reproducible fragments of customizable size (Figure 2).

The kit includes adapters that each have a unique combination of two barcodes and are compatible with all Illumina sequencers, in foil-sealed 24- or 96-well plate format for easy automation and reduced risk of cross-contamination.

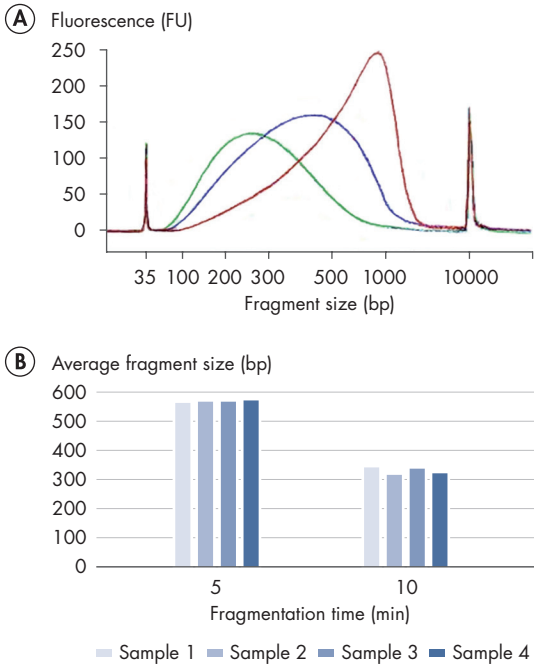

Figure 2. The QIAseq FX DNA Library Kit generates customizable, reproducible DNA fragmentation. A Fragmentation of samples using the QIAseq FX DNA Library Kit is highly customizable. Three separate libraries were produced to demonstrate the wide range of fragment sizes possible using the QIAseq FX kit. Libraries shown include inserts approximately 200 (**green**), 500 (**blue**) or 1000 (**red**) bp long. Data were generated using an Agilent® Bioanalyzer. **B** DNA samples with broad G/C contents were fragmented using the QIAseq FX DNA Library Kit with either a 5- or 10-minute run time. Fragment sizes were highly reproducible for all samples for each run time.

Low G/C bias for superior library quality

QIAseq FX technology enables library preparation with minimal G/C bias independent of the sequence context, allowing genome coverage comparable to mechanical shearing and outperforming other enzymatic methods (Figure 3). Low sequence bias also makes QIAseq FX compatible with purified input DNA from any genome, including human, plant, viral, and even bacterial species with extreme G/C content.

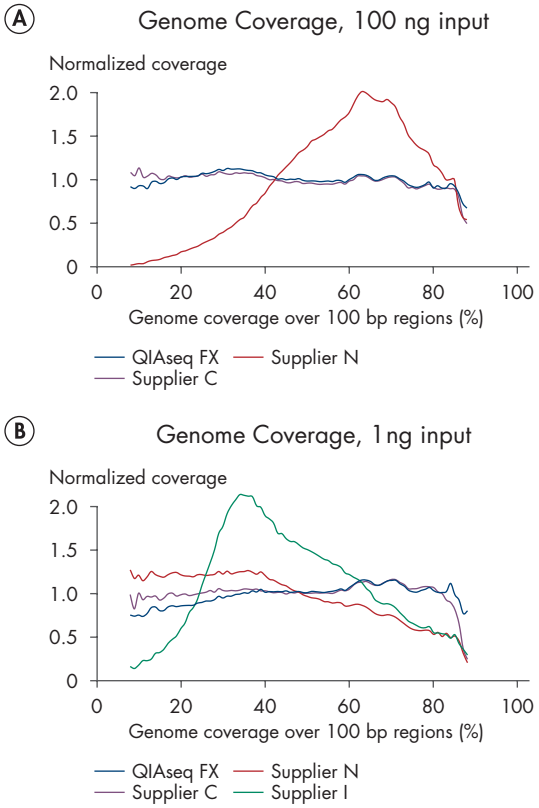

Figure 3. Minimal G/C bias compared to other enzymatic methods. Due to its highly sequence-independent fragmentation, QIAseq FX exhibits minimal G/C bias, which is comparable to mechanical shearing. **A** 100 ng genomic DNA was used as the input in comparative library preparation and sequencing. The QIAseq FX DNA Library Kit and the leading mechanical shearing method (Supplier C) produced similar and consistent coverage across a wide range of G/C contents. An all-enzymatic protocol from Supplier N yielded much higher bias toward high % G/C. **B** 1 ng genomic DNA was used as the input in a similar experiment. Again, QIAseq FX and mechanical shearing provided similar and consistent coverage, outperforming other methods.
Other supplier methods: **Supplier C**: mechanical shearing combined with a standard library prep; **Supplier N**: fragmentase; **Supplier I**: tagmentation.

Superior coverage distribution and lower duplication rate

QIAseq FX technology ensures that the majority of genomic targets have very similar total coverage depth (Figure 4). This reduces the need for additional sequencing to bring low-coverage targets up to an interpretable coverage range, saving time and resources. The duplication rate indicates the fraction of a sequencing dataset derived from PCR copies created during library preparation rather than new genomic diversity present in the original sample DNA. The same amount of sequencing from libraries with low complexity have higher duplication rates, while high-quality libraries have sufficient genomic diversity to cluster the flow cell with nearly 100% unique sequences. The QIAseq FX DNA Library Kit outperforms other enzymatic methods and gives a duplication rate similar to that achieved by mechanical shearing (Figure 4).

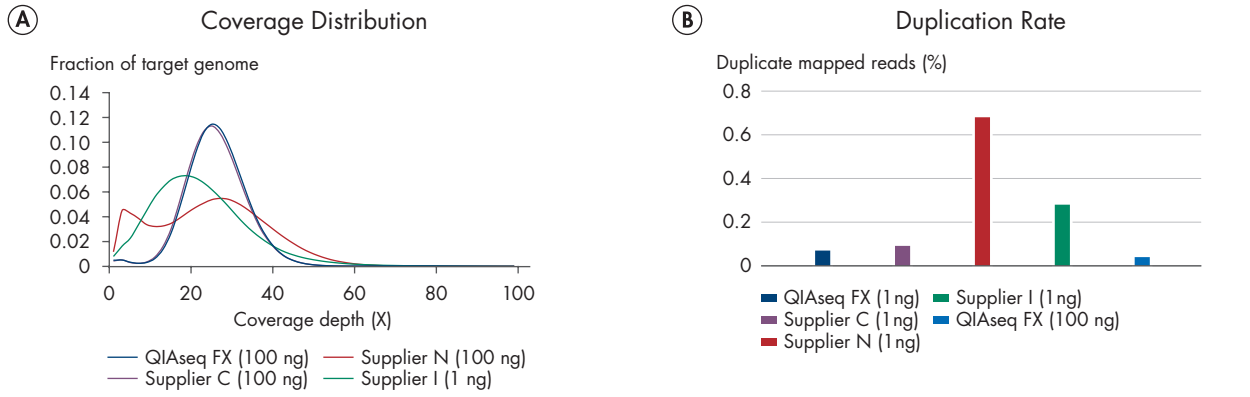

Figure 4. Superior coverage distribution and lower duplication rate. A For this comparative distribution experiment, the input was 100 ng samples of an equal-molar mixture of genomic DNA from three bacterial species with vastly different GC contents: *Fusobacterium nucleatum* with 27% GC; *Escherichia coli* with 50% GC; and *Bordetella pertussis* with 67% GC (with the exception of the sample undergoing tagmentation, as only specific input amounts are accepted). The single, narrow peak for the library created with QIAseq FX DNA Library Kit shows that the majority of genomic targets have very similar total coverage depth. This is comparable to DNA fragmented by mechanical shearing. **B** 1 ng (all kits including QIAseq FX) or 100 ng (QIAseq FX only) genomic DNA was used as the input in a comparative duplication rate experiment. At 1 ng of input, the QIAseq FX DNA Library Kit performs comparably to mechanical shearing. At 100 ng input, QIAseq FX duplication rates are even lower, approaching the level typically seen from PCR-free workflows.
Other supplier methods: **Supplier C**: mechanical shearing combined with a standard library prep; **Supplier N**: fragmentase; **Supplier I**: tagmentation.

PCR-free libraries from as little as 100 ng input

Fully functional dual-barcoded adapters and optional high-fidelity library amplification reagents enable the use of QIAseq FX fragmentation and adapter ligation chemistries to generate PCR-free libraries in under two hours total workflow time. Without additional PCR library amplification, the QIAseq FX DNA Library Kit can consistently generate the higher than 2 nM PCR-free library concentration needed for sequencing on an Illumina MiSeq or NextSeq 500 from a range of input DNA amounts (recommended amount 100 ng; lower input amounts have also been found to generate the required concentration in PCR-free workflows; Figure 5)

High-quality whole genome libraries from an all-enzymatic workflow

By providing customizable, reproducible DNA fragments from between 1 ng and 1 µg of purified genomic DNA, the QIAseq FX DNA Library Kit generates high-quality libraries with the speed and convenience of a simple, all-enzymatic workflow. QIAseq FX libraries are suitable for any whole genome or hybrid capture sequencing experiment. Dual-barcoded adapters provided in our convenient and 24- and 96-well plates also make automation easier reduce the risk of cross-contamination. Discover the flexible, customizable and high-performance QIAseq FX DNA Library Kit today!

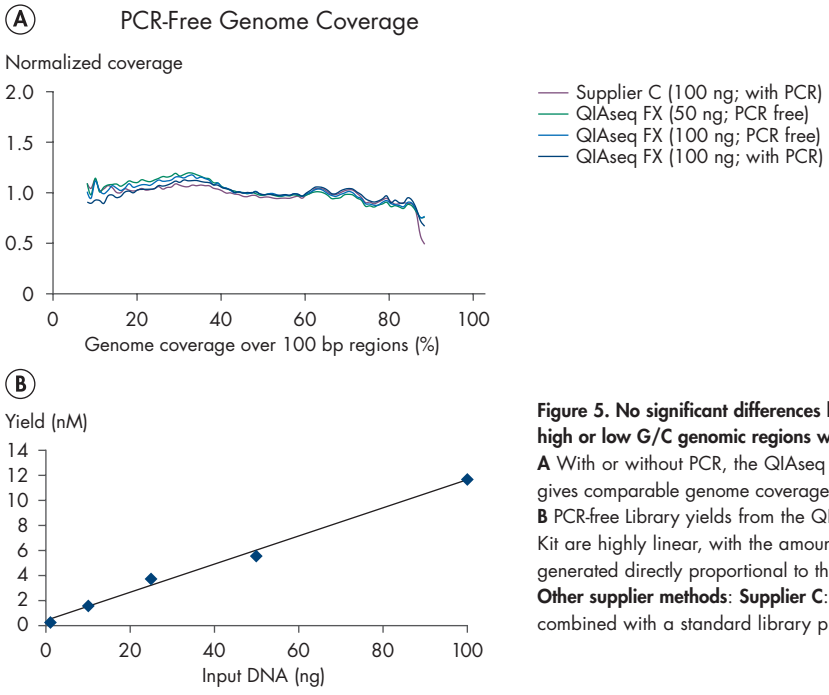

Figure 5. No significant differences between coverage of high or low G/C genomic regions with or without PCR. **A** With or without PCR, the QIAseq FX DNA Library Kit gives comparable genome coverage to mechanical shearing. **B** PCR-free Library yields from the QIAseq FX DNA Library Kit are highly linear, with the amount of total library generated directly proportional to the amount of input DNA. **Other supplier methods**: **Supplier C**: mechanical shearing combined with a standard library prep.

Ordering Information

Product	Contents	Cat. no.
QIAseq FX DNA Library Kit (96)	For 96 reactions: Buffers and reagents for DNA fragmentation, end-repair, A-addition, ligation and library amplification; for use with Illumina instruments; includes a plate containing 96 adapters with different barcodes (pierceable foil seal allowing usage of defined parts of plate)	180475
QIAseq FX DNA Library Kit (24)	For 24 reactions: Buffers and reagents for DNA fragmentation, end-repair, A-addition, ligation and library amplification; for use with Illumina instruments; includes a plate containing 24 adapters with different barcodes (pierceable foil seal allowing usage of defined parts of plate)	180473

For up-to-date licensing information and product-specific disclaimers, see the respective QIAGEN kit handbook or user manual. QIAGEN kit handbooks and user manuals are available at **www.qiagen.com** or can be requested from QIAGEN Technical Services or your local distributor.

For more information on this versatile library prep, visit **qiagen.com/qiaseq-fx**.