sparQ DNA Frag & Library Prep Kit

Reliable enzymatic fragmentation safeguards samples for sequencing success

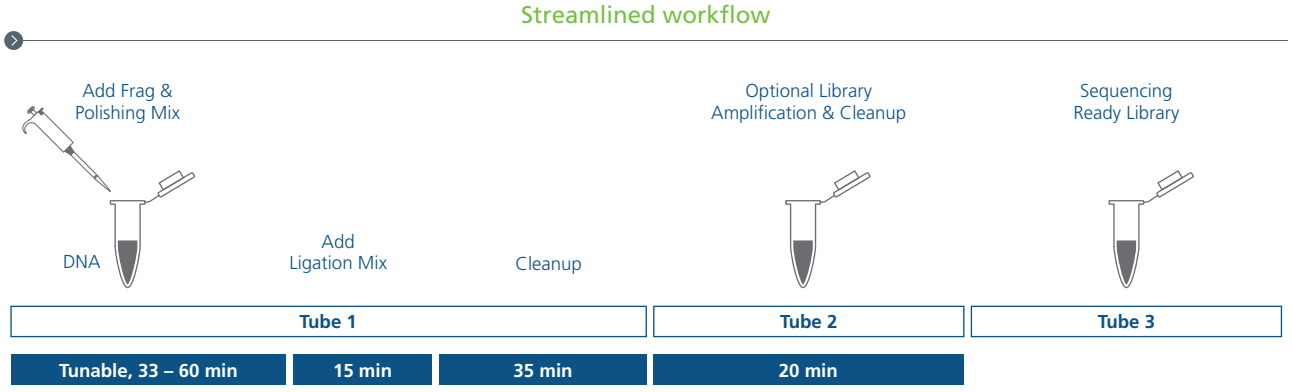
FEATURES & BENEFITS:

- Simple 2 step workflow employs a unique enzyme mix safeguarding samples from over fragmentation
- Tunable and reproducible fragmentation profiles across a range of sample types
- Flexible generation of high quality libraries from 1 ng – 1 µg of input DNA
- PCR-free workflow enabled from 100 ng
- Minimized bias across challenging regions for improved sequencing results

DESCRIPTION:

The sparQ DNA Frag & Library Prep Kit optimizes the integration of enzymatic fragmentation into a two-step protocol for the streamlined construction of libraries for sequencing on Illumina® NGS platforms. A single tube enzyme mix facilitates the combination of fragmentation and DNA polishing reactions minimizing over fragmentation while greatly simplifying library prep.

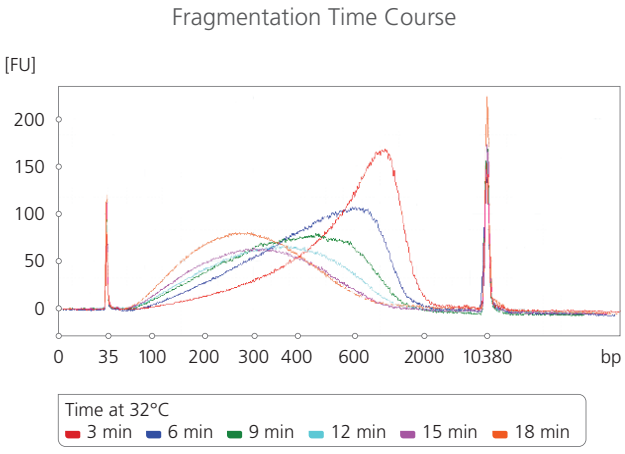
Quantabio's engineered DNA frag and polishing enzymes work in concert generating fragment sizes that are tunable and reproducible based on reaction time. Double-stranded DNA molecules are fragmented followed by DNA polishing reactions where 5'-phosphorylated and 3'-dA-tailed DNA fragments suitable for direct ligation of sequencing adapters are generated. Subsequent ligation of sequencing adapters is performed without an intervening cleanup step. The streamlined workflow can be completed in under 3 hours with minimal hands-on time accommodating DNA input amounts from 1 ng to 1000 ng. The HiFi PCR Master Mix and Primer Mix allow unbiased amplification of fragments with appropriate adapters ligated to both ends. PCR-Free workflows are enabled from 100 ng of starting material.



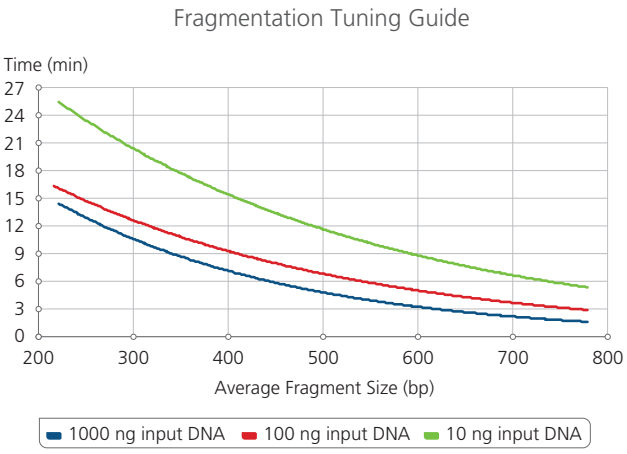
1.1 **The streamlined workflow utilizes a proprietary enzyme mix that integrates tunable and reproducible fragmentation with DNA polishing simplifying library construction and minimizing over fragmentation.** The same single reaction tube is used to proceed to adapter ligation and cleanup, minimizing sample transfer steps. A second tube is used for workflows requiring PCR amplification, and a final tube receives the sequencing-ready library.

Tunable & reproducible fragmentation

The sparQ DNA Frag & Library Prep kit is designed to produce fragments that are tunable to application specific sizes. Flexible input DNA amounts range from 1 ng - 1 µg. The single tube enzyme mix fragments DNA and then automatically proceeds to the DNA polishing reaction minimizing potential over fragmentation. sparQ DNA Frag & Library Prep kit consistently produces target fragments aligned to the desired target size.



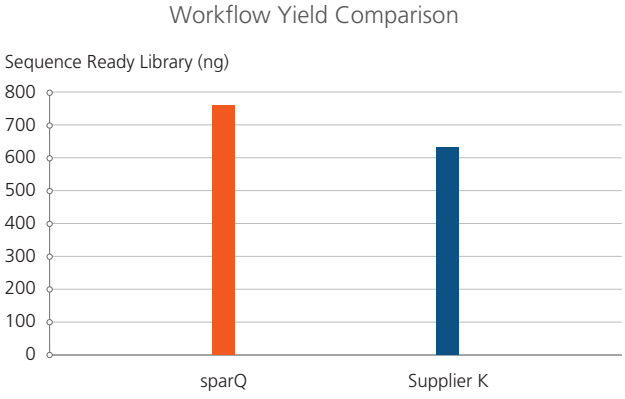
2.1 sparQ DNA Frag & Library Prep Kit is tunable to the desired fragment size. 100 ng Human gDNA was subjected to fragmentation with a series of incubation time points (3 – 18 min). After fragmentation, DNA samples were purified and then visualized using Agilent High Sensitivity DNA Kit.



2.2 Simply select the desired fragment size and input DNA amount. If input DNA falls between values displayed on the graph, an estimate can be used for optimizing fragmentation times.

Superior quality libraries & yields

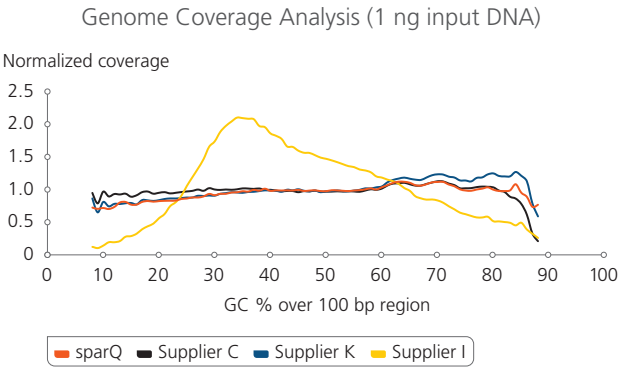
Designed to combine the highest efficiency enzymes for fragmentation, polishing, adapter ligation, and amplification, the sparQ enzymes generate superior yields of sequencing ready libraries over a broad range of input DNA. PCR-Free workflows are enabled for 100 ng input DNA. For applications requiring amplification, the high-fidelity master mix allows researchers to reduce the number of PCR cycles required to achieve the target concentration thereby reducing additional PCR-derived artifacts. Ultimately, precious samples can be conserved and reserved for additional applications when necessary.

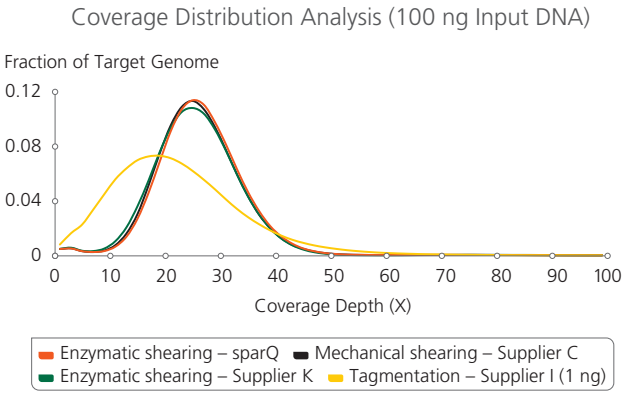


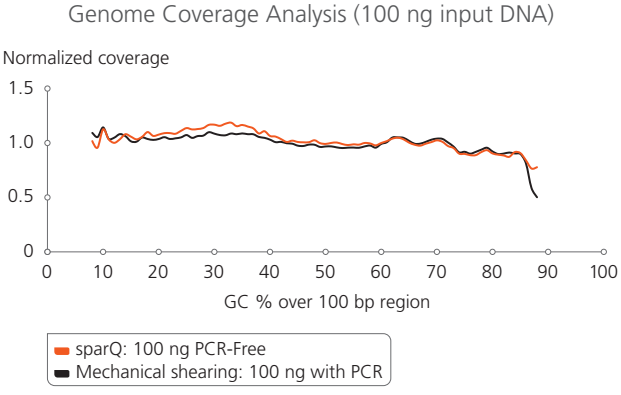
3.2 sparQ DNA Frag & Library Prep Kit shows significantly higher NGS library preparation efficiency. Libraries with 300 bp average DNA fragments from 100 ng of gDNA Coriell NA12878 were prepared using sparQ DNA Frag & Library Prep Kit and a commercial kit. Manufactures' manuals were carefully followed. Amplified libraries (5 cycles of amplification) were quantified by Qubit fluorometric quantitation method.

Minimize bias, maximize coverage uniformity to maintain genome diversity across a wide GC spectrum

The highly efficient library prep reduces coverage bias resulting in superior quality needed and expected, to minimize coverage gaps especially for challenging regions like GC- and AT-rich sequences. Reproducible and uniform genome coverage is achieved independent of input DNA amounts and closely resembles coverage obtained using mechanical shearing workflows resulting in optimized sequencing outcomes.







4.1 **Library prepared using sparQ DNA Frag & Library Prep Kit resulted in uniform coverage across the wide range of GC-content.** Libraries were prepared using different DNA fragmentation and library preparation kits with 1 ng or 100 ng of microbial gDNA followed by sequencing on Illumina MiSeq. 2 million reads from each tested library were down-sampled and analyzed. Coverage uniformity against GC-content bias resulting from different DNA fragmentation and library preparation kits were compared by plotting normalized coverage for a wide GC-content.
Libraries prepared using PCR-free workflow of sparQ DNA Frag & Library Prep Kit with 100 ng of microbial genomic DNA shows similar high performance as a typical amplified library prepared by Covaris mechanical shearing method.

Quality sequencing metrics and improved workflow economics

Optimized efficiencies in an enzymatic fragmentation 2 step protocol achieves quality sequencing metrics. The tunable fragmentation protocol safeguards samples from over fragmentation. A proprietary enzyme mix delivers high conversion efficiency and high yield of adapter ligated libraries. This allows lower input DNA amounts conserving precious sample. Finally, high fidelity amplification produces superior yields enabling fewer PCR cycles thus minimizing concerns for PCR introduced errors. The results are sequencing metrics with highly mappable reads and low duplication rates to ensure the greatest return on sequencing investment.

		1 ng input DNA		100 ng input DNA	
	Fragmentation	Mapped reads	Duplication	Mapped reads	Duplication
sparQ	Enzymatic	91.9%	0.07%	94.5%	0.04%
Supplier K	Enzymatic	92.4%	0.08%	93.5%	0.03%
Supplier I	Tagmentation	93.8%	0.28%	–	–
Supplier C	Mechanical	93.0%	0.09%	93.6%	0.03%

sparQ DNA Frag & Library Prep Kit generates high quality DNA libraries with minimal duplication artifacts. Libraries were prepared with 1 ng and 100 ng of microbial genomic DNA, amplified for 12 and 6 cycles respectively, and subsequently sequenced on Illumina MiSeq. Each library was down-sampled to 2 million reads (150 bp paired-end reads) and aligned to a reference genome with only unique alignments reported.

ORDER INFO

Product Name	Quantabio Catalog Number	Size
sparQ DNA Frag & Library Prep Kit - 24	95194-024	24 rxns
sparQ DNA Frag & Library Prep Kit - 96	95194-096	96 rxns
sparQ DNA Library Prep Kit - 24	95191-024	24 rxns
sparQ DNA Library Prep Kit - 96	95191-096	96 rxns
Related Products		
sparQ Adapter Barcode Set A	95193-A96	12 single index barcoded for 96 rxns
sparQ Adapter Barcode Set B	95193-B96	12 single index barcoded for 96 rxns
sparQ HiFi PCR Master Mix	95192-050	50 rxns (1 x 1.25 ml)
sparQ HiFi PCR Master Mix	95192-250	250 rxns (5 x 1.25 ml)
PerfeCTa® NGS Quantification Kit - Illumina - 500 R	95154-500	500 x 20 µl rxns
PerfeCTa NGS Quantification Kit - Illumina, ROX - 500 R	95155-500	500 x 20 µl rxns
PerfeCTa NGS Quantification Kit - Illumina, Low ROX - 500 R	95156-500	500 x 20 µl rxns

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