Single-tube solutions to streamline DNA library preparation and improve sequencing results while lowering costs

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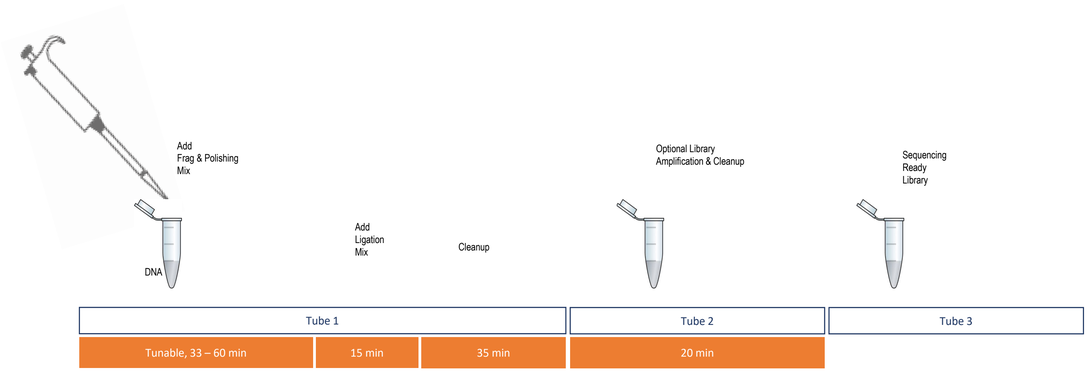
Introduction

The sparQ DNA Frag & Library Prep Kit optimizes the integration of enzymatic DNA fragmentation and library construction into a simple two-step workflow for Illumina® NGS platforms. An optimized mix of Quantabio's engineered enzymes works in concert to combine tunable DNA fragmentation and polishing reactions minimizing over fragmentation and greatly simplifying the library prep process. Resulted 5'-phosphorylated and 3'-dA-tailed DNA fragments are suitable for direct ligation of sequencing adapters 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 1 µg. The HiFi PCR Master Mix and Primer Mix allow for the unbiased amplification of DNA libraries. PCR-free workflow is enabled from 100 ng of DNA sample.

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 of input DNA
- ✓ Minimized bias across challenging regions for improved sequencing results

Library Prep Workflow



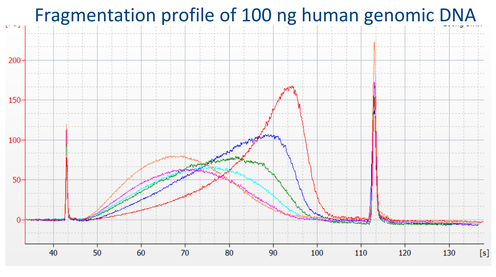
Streamlined 2-step protocol combines enzymatic DNA fragmentation and DNA Polishing (traditional end repair and a-tailing) into a single tunable step.

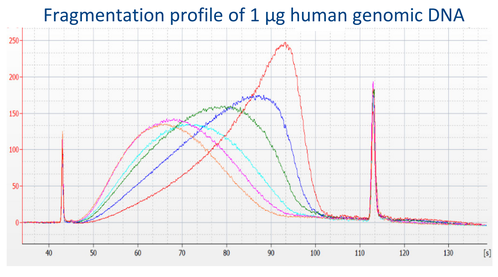
Workflow Comparisons

	NEBNext® Ultra™ II FS DNA Library Prep Kit	Nextera DNA Flex Library Prep Kit	KAPA HyperPlus Kit	sparQ DNA Frag & Library Prep Kit
Total / Hands-on Time	3 hr / 35 min	3 hr / 90 min*	2.5 hr / 35 min	2.5 hr / 30 min
Input	100 pg – 500 ng	1 – 500 ng	1 ng – 1 µg	1 ng – 1 µg**
Pros	Enzyme-based fragmentation	Tagmentation; Integrated normalization (100 – 500 ng)	Enzyme-based fragmentation	High library yield and sensitivity; Consistency; Flexibility
Cons	Low yield; Poor Reproducibility; GC Bias; Different workflows	Low yield; High duplication rate; Fixed fragment size; Different workflow for different inputs	Poor consistency and reproducibility	Optimization for different inputs
	Fragmentation, End Repair & A-tailing → Ligation → USER Excision → Purification → PCR → Purification	Tagmentation → Purification → PCR → Purification	Fragmentation → End Repair & A-tailing → Ligation → Purification → PCR → Purification	Fragmentation & End Polishing → Ligation → Purification → PCR → Purification

* vendor specified time ** down to 100 pg

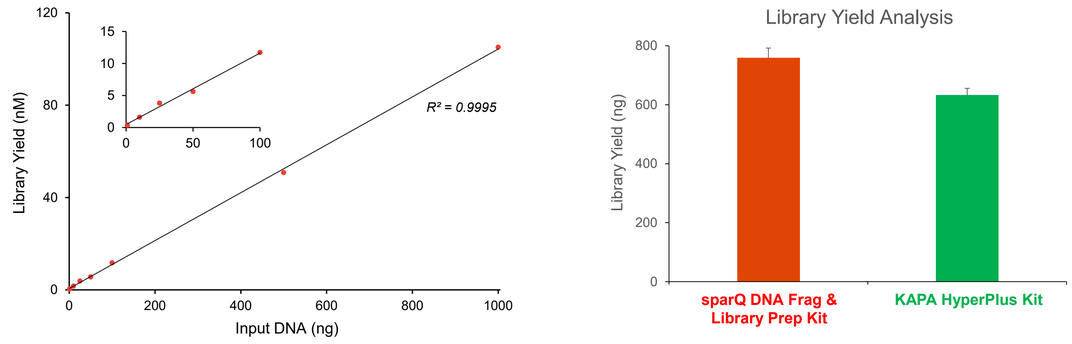
Tunable Size Ranges for Varying DNA Inputs (1 ng – 1 µg)





sparQ DNA Frag & Library Prep Kit is designed to produce DNA fragments which are tunable to specific sizes for different applications. 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. The sparQ DNA Frag & Library Prep Kit consistently produces target fragments aligned to the desired target size.

Superior Efficiency & Yields



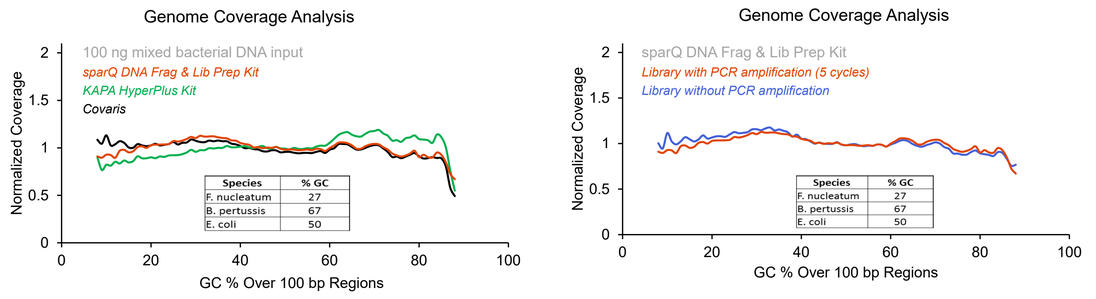
Excellent dynamic range indicates consistent library prep efficiency across a wide range of input DNA (1 ng – 1 µg)

High Quality DNA Libraries

	Fragmentation Method	1 ng Input DNA		100 ng Input DNA	
		Mapped Reads*	Duplication	Mapped Reads*	Duplication
sparQ DNA Frag & Lib Prep Kit	Enzymatic	91.92%	0.07%	94.45%	0.04%
KAPA HyperPlus Kit	Enzymatic	92.40%	0.08%	93.49%	0.03%
Covaris	Mechanical	92.96%	0.09%	93.61%	0.03%

sparQ DNA Frag & Library Prep Kit produces high quality libraries with high read alignment percentages and low duplication rates

Uniform Coverage Across Challenging Genomic Regions



DNA libraries prepared by sparQ DNA Frag & Library Prep Kit with either library amplification or PCR-free workflow maintain even coverage across extreme AT- and GC-rich regions of the genome

sparQ Compared to Other Technologies

Conclusions

sparQ DNA Fragmentation & Library Prep Kit

- Enzyme Mix integrates fragmentation and DNA polishing reactions for a streamlined workflow
- Complete workflow in under 3 hours with minimal hands-on time
- Adjusting fragmentation time generates selective DNA sizes
- Low fragmentation and amplification bias enables more uniform genome coverage
- Excellent sequencing metrics
- Capable of PCR-free library prep workflow