



Evolve LIBRARY PREP

KAPA HyperPlus Kit. NGS library preparation. Evolved.

The KAPA HyperPlus Kit provides a streamlined workflow in which DNA fragmentation and library construction are performed in a single tube. This integrated solution combines the industry-leading library construction efficiency and library quality of the KAPA HyperPrep Kit with the speed and convenience of enzymatic fragmentation.

KAPA HyperPlus Kits offer complete library preparation solution with KAPA Adapters and KAPA HyperPure Beads (sold separately). Validated with the KAPA HyperCap and KAPA HyperPETE workflows.

Benefits of the KAPA HyperPlus Workflow

Exceptional performance and sequencing results

- Industry-leading conversion rates (library prep efficiency) and library complexity, particularly for FFPE DNA
- Reduced bias and more uniform sequence coverage
- Includes an additional End Repair and A-Tailing module for more sensitive applications with no change to the workflow

Tunable and trusted

- Compatible with a wide range of sample types and inputs, and flexible with respect to fragment size, adapter design and library amplification

Automation friendly

- PCR-free workflows from lower inputs
- Qualified automation methods



Integrated fragmentation and library preparation solution

The KAPA HyperPlus Kit includes low-bias enzymatic fragmentation, eliminating the need for mechanical DNA shearing methods that require expensive instrumentation and are difficult to automate.

- Fragment DNA and construct libraries in 2.5 hours with a single-tube, automation-friendly workflow
- High success rates from a wide range of DNA input amounts and sample types, including challenging samples such as FFPE
- Suitable for a variety of sequencing applications, including human exome and microbial whole genome sequencing

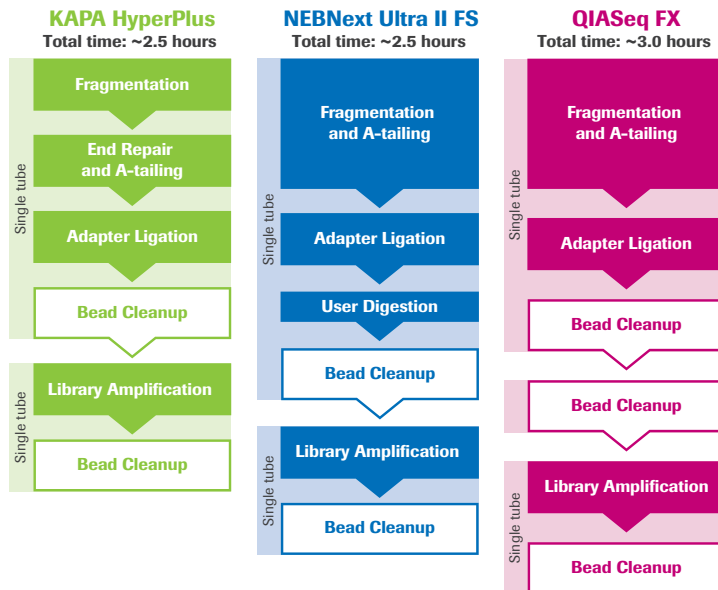


Figure 1. The KAPA HyperPlus workflow provides a streamlined workflow that includes fully automatable fragmentation and library preparation in a single tube.

Tunable and reproducible fragmentation

- Adjust library insert sizes from 150 – 800 bp by varying fragmentation time
- Reproducible insert sizes across a range of GC content and DNA input amounts

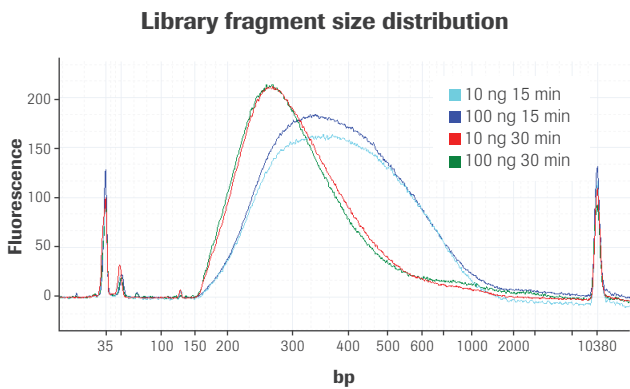


Figure 2. Reproducible library fragment size distributions are obtained with different DNA inputs. Various input amounts of *Escherichia coli* gDNA were processed using the KAPA HyperPlus Kit with fragmentation times of 15 or 30 minutes at 37°C. After library amplification and a single bead cleanup, samples were analyzed using an Agilent® High Sensitivity DNA Assay.

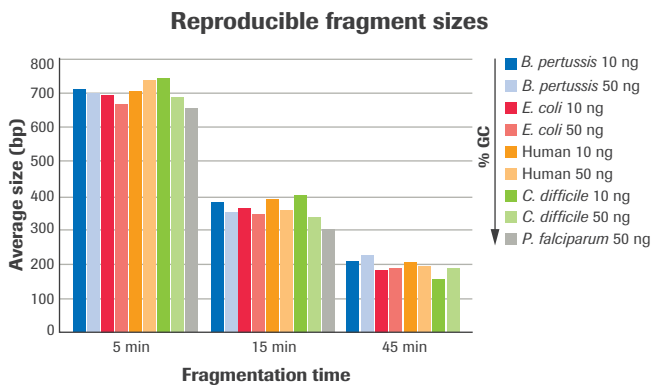


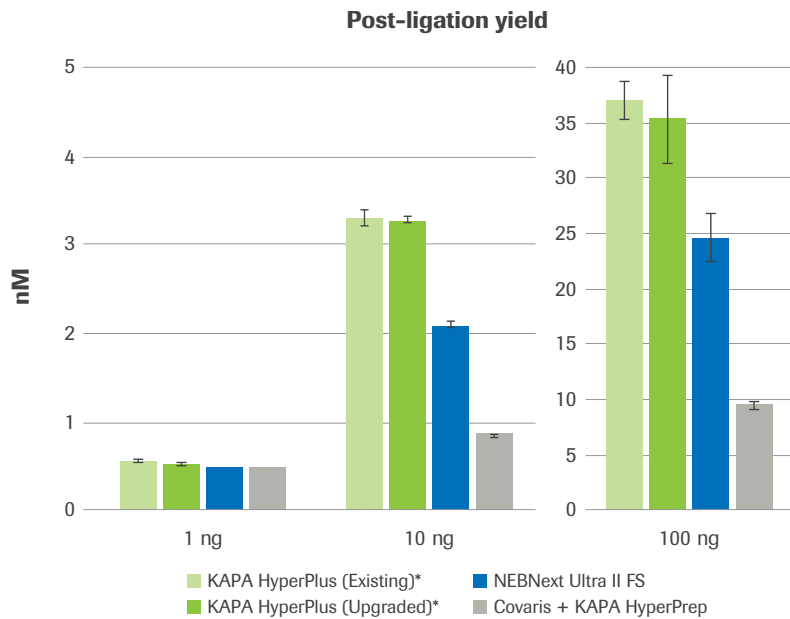
Figure 3. Defined fragmentation parameters yield consistent library insert sizes for samples from multiple species across a wide range of GC content. 10 ng or 50 ng of *Bordetella pertussis* (68% GC), *Clostridium difficile* (29% GC), *Escherichia coli* (51% GC), *Plasmodium falciparum* (20% GC) or human gDNA were fragmented for 5, 15, or 45 minutes. These fragmentation times yielded average library insert sizes of approximately 700 bp, 350 bp and 200 bp, respectively; irrespective of GC content and input amount. All fragmentation reactions were performed at 37°C.

Industry-leading library yields

High conversion rates, defined as % input DNA converted to sequenceable, adapter-ligated library, lead to higher PCR-free yields, which ultimately determines library diversity and quality.

- Low-input samples no longer require specialist library construction workflows or reagents
- PCR-free workflows from as little as 50 ng starting material
- Both the existing and upgraded KAPA HyperPlus chemistry produce the highest post-ligation yields

Figure 4. KAPA HyperPlus Kits produce the highest yields from a range of input amounts. The KAPA HyperPlus Kit (both existing and upgraded workflows) converts more input DNA to adapter-ligated library than NEBNext® Ultra II FS Kits or Covaris shearing combined with KAPA HyperPrep. PCR-free libraries were prepared from 1 ng, 10 ng, and 100 ng *E. coli* gDNA using KAPA UDI Adapters and KAPA HyperPure Beads for all workflows, including NEBNext Ultra II FS Kits. Libraries were quantified after ligation using the KAPA Library Quantification Kit.



Enable superior sequencing results

- Higher post-ligation yields result in fewer amplification cycles and lower duplication rates
- Detect low-frequency mutations with high confidence due to decreased sequencing artifacts and increased sensitivity with the upgraded version of the KAPA HyperPlus Kit

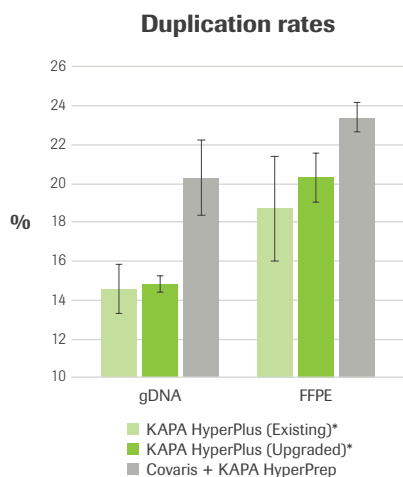


Figure 5. Lower duplication rates. Whole human exome libraries were prepared using 100 ng inputs with the existing and upgraded KAPA HyperPlus Kit, or the KAPA HyperPrep Kit. Captures were performed with the SeqCap EZ MedExome panel.

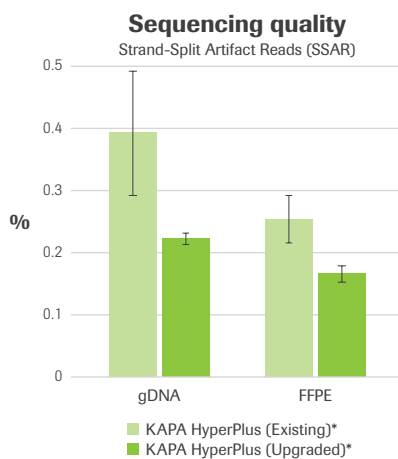


Figure 6. Improved sequencing performance. Whole human exome libraries were prepared using 100 ng inputs with the existing and upgraded KAPA HyperPlus Kit. Captures were performed with the SeqCap EZ MedExome panel. Strand-split artifact reads (SSARs) represent chimeric reads that appear to be derived from non-contiguous portions of the genome.¹

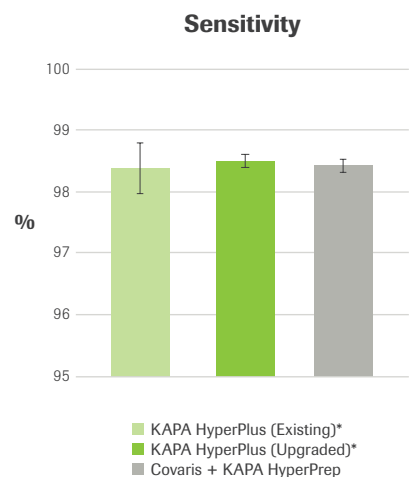


Figure 7. Higher single nucleotide polymorphism (SNP) sensitivity. Whole human exome libraries were prepared using 100 ng high-quality gDNA with the KAPA HyperPrep Kit, the existing and upgraded KAPA HyperPlus Kit. Captures were performed with the SeqCap EZ MedExome panel.

*KAPA HyperPlus (existing) represents the use of the HyperPrep End Repair & A-Tailing Enzyme Mix (original enzyme); KAPA HyperPlus (upgraded) represents the use of the HyperPlus End Repair & A-Tailing Enzyme Mix (a new enzyme that is now included in the KAPA HyperPlus Kit).

Minimal sequence coverage bias

- Lower sequence bias when compared other enzymatic fragmentation methods
- Less bias leads to more uniform sequencing coverage and reduced sequencing costs

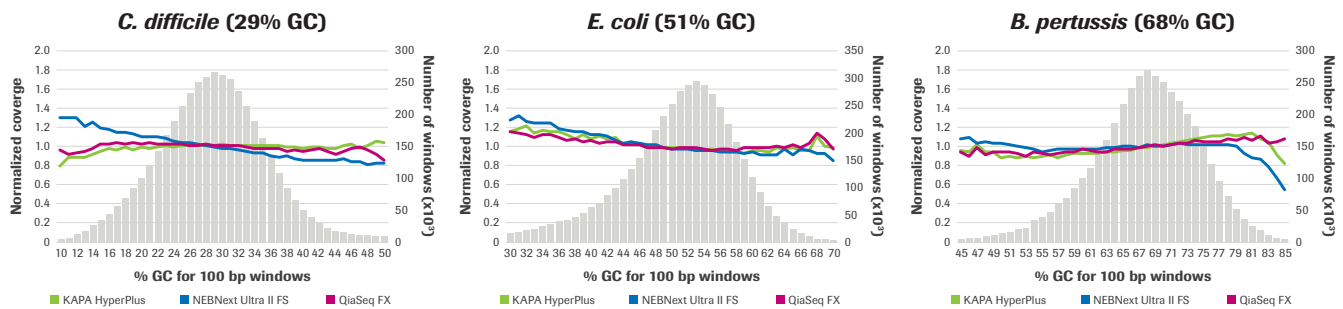


Figure 8. GC bias comparison. GC bias for *C. difficile* (left), *E. coli* (middle), and *B. pertussis* (right) was assessed by calculating the GC content of the reference in 100 bp bins and plotting normalized coverage across these bins for the KAPA HyperPlus, NEBNext® Ultra™ FS, and QiaSeq FX workflows, using Picard CollectGCBiasMetrics. Libraries were prepared from 10 ng of input DNA. In the absence of sequencing bias, all bins would be equally represented, indicated by a horizontal distribution centered on a normalized coverage of 1. Distribution of GC content in the genome is indicated by the grey histograms.

Ordering information

Roche cat. no.	KAPA code	Description	Pack size
07962380001	KK8510	KAPA HyperPlus Kit with Library Amplification	8 reactions
07962401001	KK8512	KAPA HyperPlus Kit with Library Amplification	24 reactions
07962428001	KK8514	KAPA HyperPlus Kit with Library Amplification	96 reactions
07962398001	KK8511	KAPA HyperPlus Kit, PCR-free	8 reactions
07962410001	KK8513	KAPA HyperPlus Kit, PCR-free	24 reactions
07962436001	KK8515	KAPA HyperPlus Kit, PCR-free	96 reactions

*Source: Haile, et al. (2019) Sources of erroneous sequences and artifact chimeric reads in next generation sequencing of genomic DNA from formalin-fixed paraffin-embedded samples. *Nucleic Acids Research*, 2019, 47,2. doi: 10.1093/nar/gky1142.

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