



KAPA HYPER PLUS NGS library preparation. Evolved.



The KAPA HyperPlus kit provides a streamlined workflow that includes fragmentation and library preparation in a single tube. Building on industry-leading library construction efficiencies, this integrated solution combines enzymatic fragmentation, similar in quality to mechanical shearing, with the speed and convenience of tagmentation-based workflows.

Benefits include:

- DNA fragmentation and library prep in 2.5 hours
- flexible DNA sample input from 1 ng 1 ug
- reduced bias and more uniform sequence coverage
- PCR-Free workflows

Integrated Fragmentation and Library Preparation Solution

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

- Fragment DNA and construct libraries in 2.5 hours with a single-tube workflow
- Supports a wide range of DNA types and input amounts including challenging sample types such as FFPE
- Versatile kit works across many applications, including human exome and microbial whole genome sequencing
- Automation-friendly workflow



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



Reproducible library fragment size distributions are obtained from a variety of DNA sample inputs. Various input amounts of *E. coli* gDNA, were processed using the HyperPlus Kit with fragmentation times of 15 or 30 minutes at 37°C. After library amplification and a single 1x SPRI[®] bead cleanup, samples were analysed using an Agilent High Sensitivity DNA Assay[™].

Reproducible Fragment Sizes



Defined fragmentation parameters yield consistent library insert sizes using 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, yielding average insert fragment sizes of ~700, ~350 or ~200 bp respectively. Fragmentation reaction performed at at 37°C.

Industry Leading Library Yields...

Conversion rate, defined as % input DNA converted to sequenceable, adapter-ligated library, is a key library construction metric, which ultimately determines library diversity and quality.

- Attain up to 100% conversion rates
- Superior performance across a range of DNA input amounts
- High library yields enable PCR-free workflows from as little as 50 ng starting material

Conversion Rate Ranges



Conversion rates vary widely for commercial library construction products. The HyperPlus integrated workflow results in greater conversion of input DNA to adapterligated library compared to Covaris-sheared DNA processed using the Illumina TruSeq Nano Prep Kit, or the KAPA Hyper Prep Kit. Conversion rates are highest for the KAPA HyperPlus Kit for both high- and low-input applications.

... Enable Superior Sequencing Results

- Higher conversion rates results in fewer amplification cycles and lower duplication rates
- Detect low-frequency mutations with high confidence due to greater library diversity and more uniform sequence coverage



Sensitivity of SNP Detection



Low duplication rates. % duplicates from exome sequencing experiments using HyperPlus, or Covaris and Hyper Prep. Libraries were prepared from 50 ng hgDNA or 50 ng FFPE DNA and captured with the Nimblegen SeqCap EZ HGSC VCRome panel.

Single nucleotide polymorphism (SNP) detection sensitivity indicates better sequencing quality. Libraries prepared for capture using the Nimblegen SeqCap EZ HGSC VCRome panel, 50 ng input, standard capture protocol. SNPs were called using LoFreq with

protocol. SNPs were called using LoFreq with default settings and a minimum per-base depth of coverage cut-off of 10. Sensitivity (true positive rate) was calculated as the ratio of true positives divided by the sum of true positives and false negatives. The total number of SNPs analyzed was greater than 25,000.

Sequence Coverage Uniformity



Sequence coverage uniformity comparison.

Data for all libraries were down-sampled to equal number of reads. Roche Nimblegen capture libraries prepared with HyperPlus or Covaris and Hyper Prep were compared to libraries prepared with Nextera™ Rapid Capture. Coverage distribution was similar for Covaris and HyperPlus libraries, while Nextera libraries displayed much broader coverage distribution. A sharper peak and smaller tails indicate more uniform coverage.

Minimal Sequence Coverage Bias

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



GC bias comparison. GC bias for C. difficile (left), E.coli (middle) 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 Covaris and Hyper Prep, HyperPlus, NEBNext[®] Ultra™ and Nextera™ workflows, using Picard CollectGCBiasMetrics. Libraries were prepared from 1 ng of input DNA. In the absence of sequencing bias, all bins would be equally represented, indicated by a horizontal distribution centred on a normalized coverage of 1. Distribution of GC content in the genome is indicated by the grey histograms.

Ordering Information

Kit Code	Description	Kit Size	
KK8510	KAPA HyperPlus Kit with Library Amplification	8 reactions	Contact Sales by: Calling us at 781.497.2933
KK8512	KAPA HyperPlus Kit with Library Amplification	24 reactions	Visiting our website at kapabiosystems.com
KK8514	KAPA HyperPlus Kit with Library Amplification	96 reactions	emailing us at sales@kapabiosystems.com
KK8511	KAPA HyperPlus Kit, PCR-free	8 reactions	
KK8513	KAPA HyperPlus Kit, PCR-free	24 reactions	Contact Technical Support by:
KK8515	KAPA HyperPlus Kit, PCR-free	96 reactions	Empiling us at support@kapabiosystems.com



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