KAPA RNA HyperPrep Kits

Single-day RNA

The KAPA RNA HyperPrep Kits utilize novel chemistry that enables the combination of enzymatic steps and fewer reaction purifications, resulting in a truly streamlined solution for the preparation of high-quality RNA-seq libraries. The strand-specific workflow is flexible, supporting library construction from lower-input amounts and degraded samples, and is compatible with both mRNA capture and ribosomal depletion. Kits contain all reagents required for RNA enrichment (if performed) and library preparation, with the exception of KAPA Adapters (available separately).

Benefits:

- Single-day library construction, inclusive of RNA enrichment
- Higher success rates with low-input and degraded samples
- Robust performance across different sample types and input amounts
- Qualified automation methods
- Complete library prep solution with KAPA Pure Beads (included) and KAPA Adapters

KAPABIOSYSTEMS

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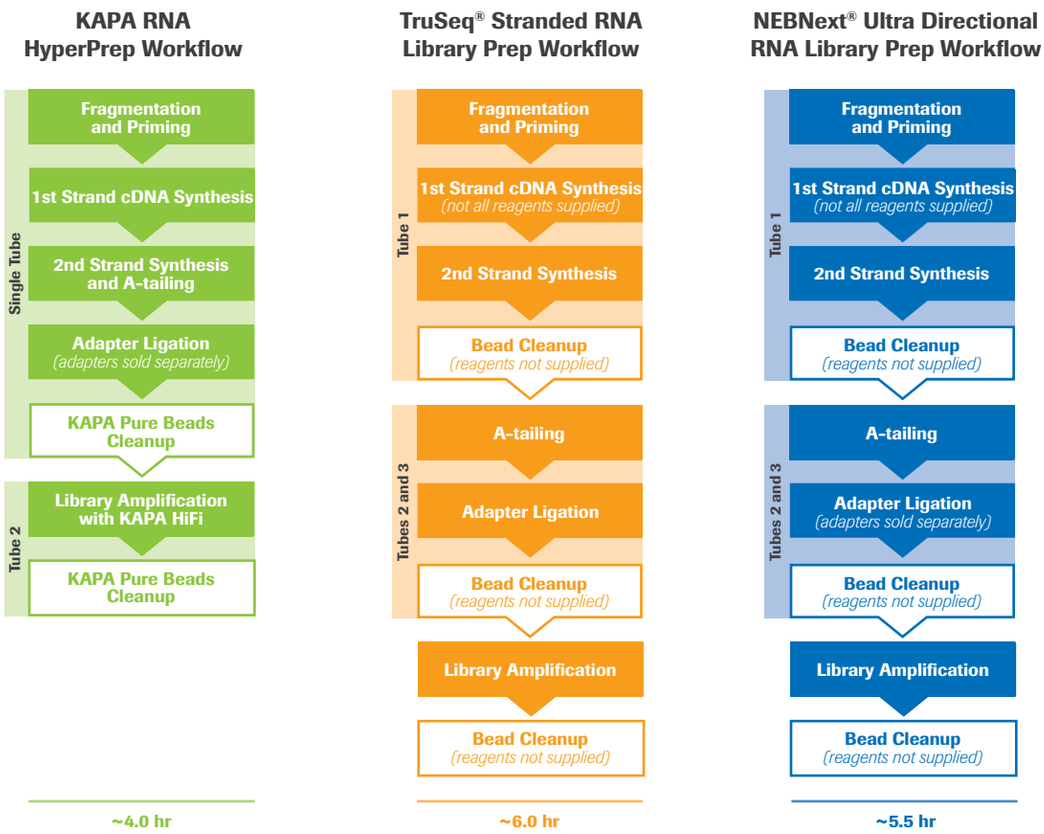
Single-tube, single-day library prep

- Reduce hands-on and overall time through fewer enzymatic and reaction cleanup steps
- Produce strand-specific, sequencing-ready libraries from input RNA in approximately 4 hours
- Complete entire workflow, inclusive of mRNA capture or ribosomal depletion, in a standard workday
- Achieve high throughput and consistency with an automation-friendly workflow



Streamlined RNA library preparation. The KAPA RNA HyperPrep workflow reduces overall library preparation time by 1.5 to 2 hours, in comparison to competitor workflows, making library construction possible in a single workday. Additionally, the reduction in the total number of enzymatic and reaction cleanup steps reduces the hands-on time required.

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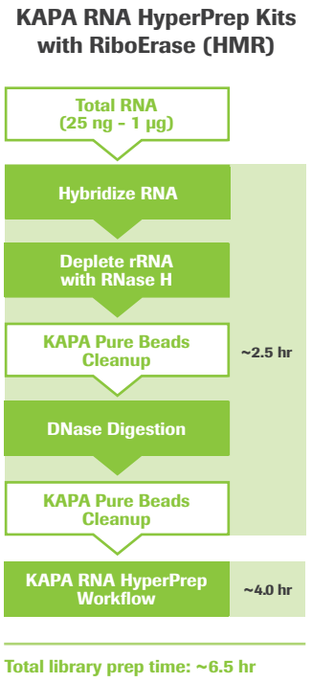
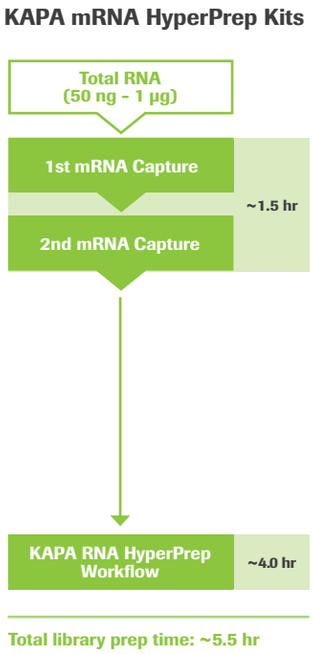
Flexible workflow options

- Use the KAPA RNA HyperPrep Kit as a standalone workflow, or combine with either the mRNA capture or KAPA RiboErase (HMR) ribosomal RNA depletion modules





KAPA RiboErase (HMR). Sequencing of rRNA-depleted total RNA samples provides a more comprehensive representation of the whole transcriptome. rRNA is targeted and depleted enzymatically using DNA probes and RNase H resulting in improved coverage of transcripts of interest, including precursor mRNAs and important regulatory species, such as noncoding RNAs.

mRNA Capture. Sequencing of mRNA-enriched samples provides a focused view of the protein-coding regions in the transcriptome. mRNA capture beads are used prior to library preparation with the KAPA RNA HyperPrep workflow, which enriches for mRNA over non-polyadenylated species, such as ribosomal, precursor, and noncoding RNAs.

Enable a variety of strand-specific applications

- Input less starting material than other commercially-available workflows
- Generate high-quality libraries, even with degraded samples, such as FFPE

	KAPA RNA HyperPrep Kits	KAPA RNA HyperPrep Kits with RiboErase (HMR)	KAPA mRNA HyperPrep Kits
RNA Enrichment	None	rRNA Depletion	Poly(A) Selection
Input Amount	1 – 100 ng into library prep	25 ng – 1 µg into rRNA depletion	50 ng – 1 µg into mRNA capture
Sample Type	High-quality total RNA Degraded or FFPE total RNA Previously enriched RNA	High-quality total RNA Degraded or FFPE total RNA	High-quality total RNA
Species	Eukaryotic (animal, plant, etc.) Prokaryotic (bacterial, etc.)	Human, mouse, and rat	Eukaryotic (animal, plant, etc.)
Differentiating Applications	Whole transcriptome	Non-coding RNA Whole transcriptome	mRNA-Seq
Shared Applications	Gene expression analysis; detection of gene fusions, isoforms, and other structural variants; novel transcript identification; SNV discovery		
Strand-specific	Yes	Yes	Yes
Automation-friendly	Yes	Yes	Yes

A workflow to meet a variety of needs. The KAPA RNA HyperPrep workflow is available in three formats: with mRNA capture, with KAPA RiboErase (HMR) for rRNA depletion, or with no RNA enrichment reagents. This flexibility allows users to select the workflow that best meets the needs of their specific application.

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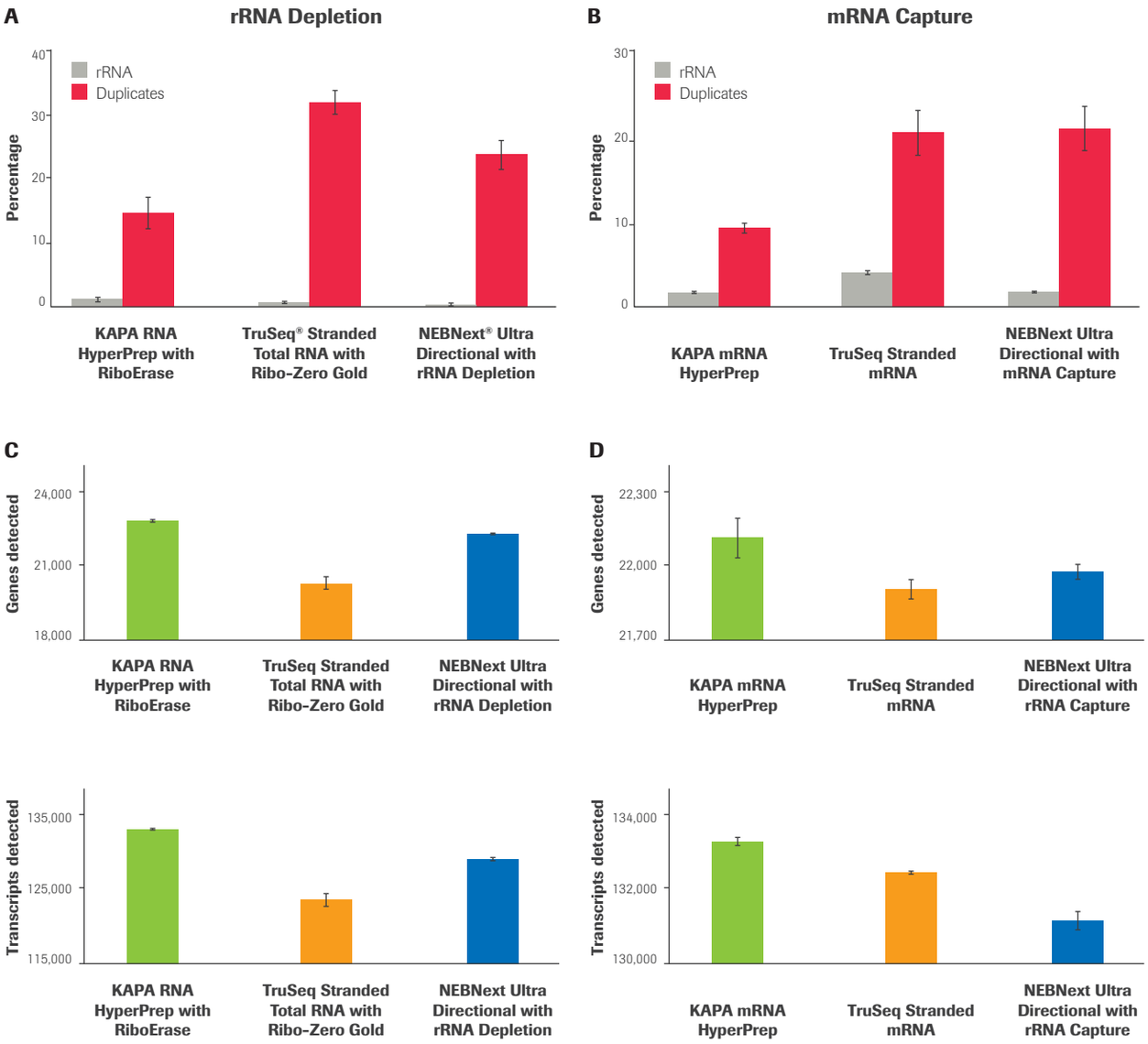
Sequence what matters

- Waste fewer reads due to the combination of rRNA carryover and PCR duplicates
- Identify more unique transcripts and genes with equivalent sequencing



Better utilize sequencing capacity. The KAPA RNA HyperPrep workflows result in a reduction in the total number of reads wasted due to both PCR duplicates and alignments to rRNA loci **(A** and **B)**. With an equivalent amount of sequencing, more genes and unique transcripts are identified using the KAPA workflows in comparison to the TruSeq® and NEBNext® kits **(C** and **D)**. Libraries were generated in quadruplicate from 25 ng (rRNA depletion) and 50 ng (mRNA capture) of high-quality Universal Human Reference (UHR) RNA using the manufacturers' standard recommendations per workflow, where possible. For this and all subsequent data, sequencing was performed using an Illumina® HiSeq® 2500 in high output mode with v4 chemistry and 2 x 100 bp read length. Reads aligning to rRNA were removed and paired reads were randomly subsampled to 14M for comparative analyses, including marked duplicates.

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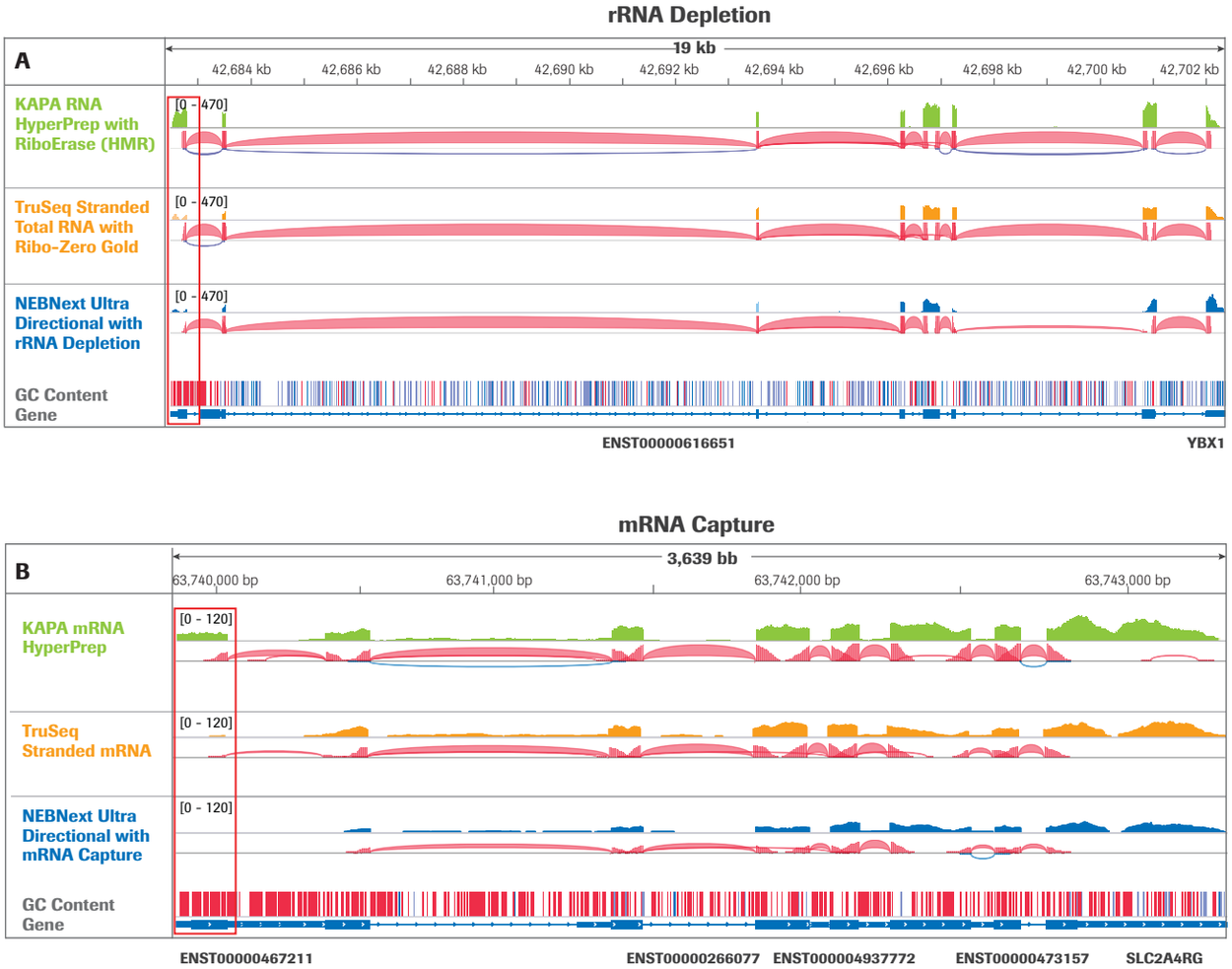
Achieve superior coverage uniformity

- Obtain more uniform distribution of reads across transcripts
- Improve coverage of difficult GC-rich regions



Improved coverage uniformity. Increased coverage of GC-rich regions (outlined in red) of the YBX1 **(A)** and SLC2A4RG **(B)** genes is demonstrated using KAPA RNA HyperPrep workflows. For the top 1000 transcripts, the KAPA workflows resulted in more even coverage across transcript lengths than competitors, as assessed by both a normalized coverage plot and coverage coefficient of variation (CV), **(C** and **D)**. Libraries were generated from 25 ng (rRNA depletion) and 50 ng (mRNA capture) of high-quality UHR RNA using the manufacturers' standard recommendations per workflow, where possible.

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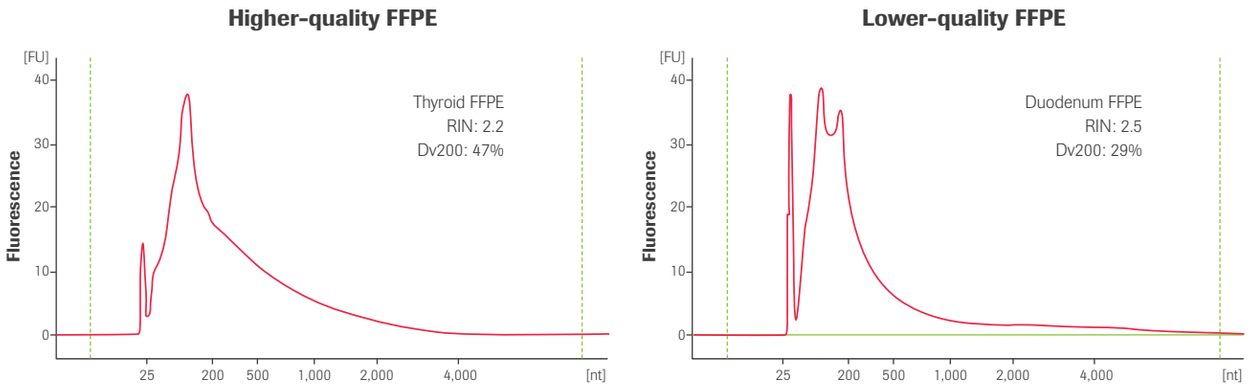
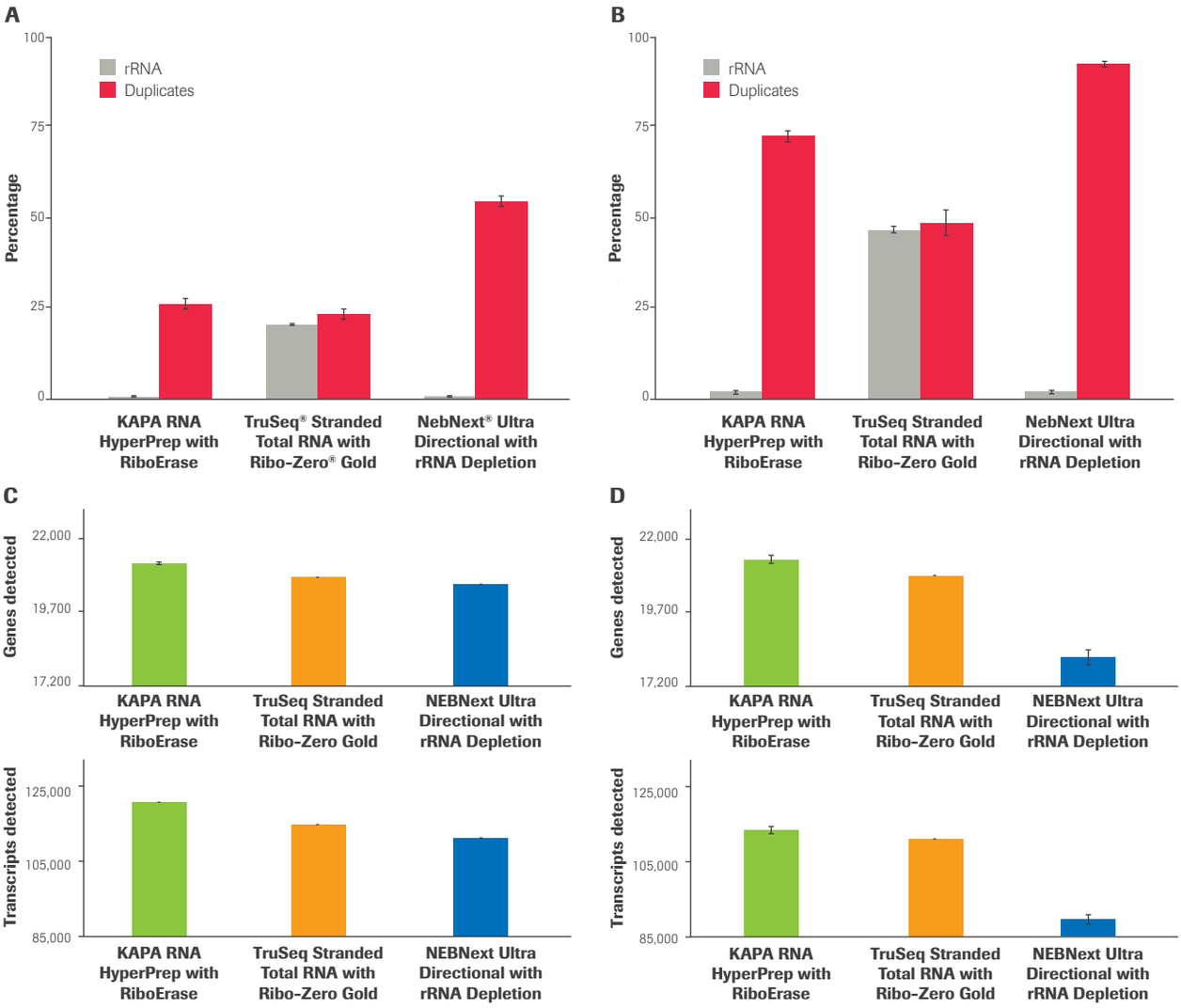
Generate high-quality libraries from degraded samples

- Prepare libraries with the KAPA RNA HyperPrep Kit with RiboErase from as low as 25 ng FFPE RNA, depending on total RNA quality
- Achieve low duplication rates and highly efficient, reproducible rRNA removal with degraded samples
- Identify more unique transcripts and genes with equivalent sequencing



Total RNA electropherograms for two FFPE samples. The thyroid FFPE sample (RIN: 2.2) is of higher quality, with 47% of the RNA measuring >200 nt. In contrast, the duodenum FFPE sample (RIN: 2.5) is of lower quality, with 29% of the RNA measuring >200 nt. Electropherograms were generated using an Agilent® RNA 6000 Pico Kit.



Better utilize sequencing capacity. Using the two FFPE RNA samples shown above, the KAPA RNA HyperPrep Kit with RiboErase (HMR) results in a reduction in the total number of reads wasted due to both PCR duplicates and alignment to rRNA loci in comparison to competitor workflows **(A** and **B)**. With equivalent sequencing, more genes and transcripts are identified using the KAPA workflow in comparison to competitors **(C** and **D)**. Libraries were generated in duplicate from 25 ng for thyroid libraries and a minimum of 100 ng for duodenum libraries, due to the lower quality of the duodenum starting material.

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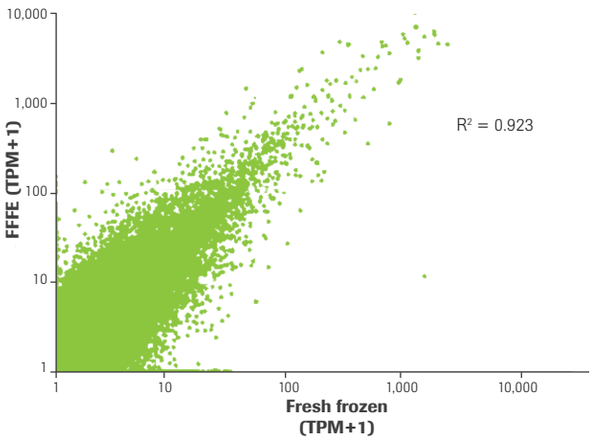
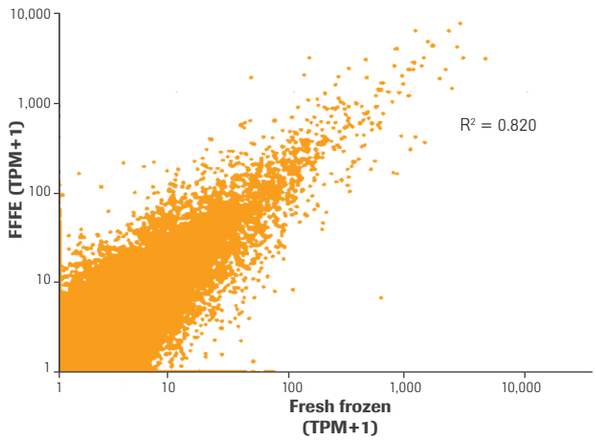
Achieve reliable results with degraded inputs

- A high degree of expression correlation between paired FFPE and fresh frozen samples, improves confidence in sequence data accuracy

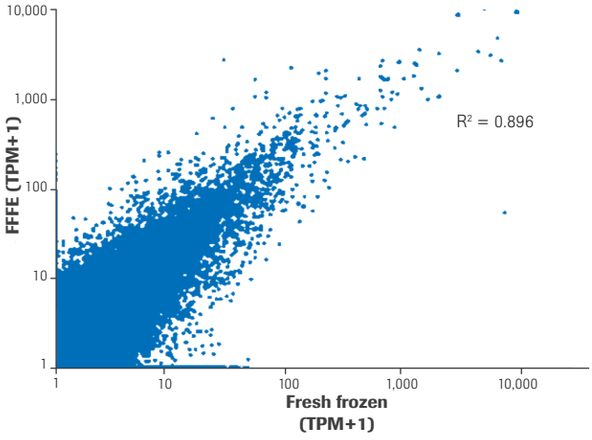
KAPA RNA HyperPrep Kit with RiboErase (HMR)



TruSeq Stranded Total RNA Library Prep Kit with Ribo-Zero Gold



NEBNext Ultra Directional Library Prep Kit with rRNA Depletion



High level of agreement between paired FFPE and fresh frozen expression data, in transcripts per million (TPM). Pearson correlation coefficients show a higher degree of agreement with the KAPA RNA HyperPrep Kit with RiboErase (HMR), in comparison to the TruSeq® Stranded Total RNA Library Prep with Ribo-Zero Gold® and NEBNext® Ultra Directional Library Preparation with the rRNA Depletion Kits. Libraries were generated in duplicate from 100 ng inputs of paired FFPE-derived and fresh frozen breast tumor total RNA samples, using the manufacturers' standard recommendations per workflow.

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Ordering Information for KAPA RNA HyperPrep Kits

Roche Cat. No.	KAPA Code	Description	Kit Size
08098093702	KK8540	KAPA RNA HyperPrep Kit	24 rxn
08098107702	KK8541	KAPA RNA HyperPrep Kit	96 rxn
08098115702	KK8580	KAPA mRNA HyperPrep Kit	24 rxn
08098123702	KK8581	KAPA mRNA HyperPrep Kit	96 rxn
08098131702	KK8560	KAPA RNA HyperPrep Kit with RiboErase (HMR)	24 rxn
08098140702	KK8561	KAPA RNA Hyper Prep Kit with RiboErase (HMR)	96 rxn

Ordering Information for KAPA Adapters

Roche Cat. No.	KAPA Code	Description	Kit Size
08005699001	KK8700	KAPA Single-Indexed Adapter Kit, Set A + B (30 µM)	24 adapters x 40 µL each
08005702001	KK8701	KAPA Single-Indexed Adapter Kit, Set A (30 µM)	12 adapters x 40 µL each
08005729001	KK8702	KAPA Single-Indexed Adapter Kit, Set B (30 µM)	12 adapters x 40 µL each
08005770001	KK8710	KAPA Single-Indexed Adapter Kit, Set A + B (1.5 µM)	24 adapters x 40 µL each
08005788001	KK8711	KAPA Single-Indexed Adapter Kit, Set A (1.5 µM)	12 adapters x 40 µL each
08005796001	KK8712	KAPA Single-Indexed Adapter Kit, Set B (1.5 µM)	12 adapters x 40 µL each

Published by:

Roche Sequencing Solutions, Inc.
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Pleasanton, CA 94588

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SEQ100007 A038 02/17