WATCHMAKER GENOMICS

WATCHMAKER mRNA LIBRARY PREP KITS

Watchmaker mRNA Library Prep Kits generate stranded mRNA-seq libraries in under 5 hours with fewer handling and cleanup steps. Designed with automation in mind, kits come with generous overages and leverage on-bead washing steps during poly(A) selection to reduce resuspension and wash times as well as consumable use.

Use a single, flexible workflow to process a broad total RNA input range (2.5 ng to 1 µg), without the need for specialized low-input solutions that fail to maintain strand origin information. The Watchmaker mRNA Library Prep Kit delivers highly complex libraries for superior gene detection sensitivity — even with challenging low-input samples.

KEY FEATURES & BENEFITS

- Construct **stranded** mRNA libraries in **under 5 hours** – enabling completion in a single workday
- **Improve library complexity** and gene detection sensitivity with challenging low-input samples **(down to 2.5 ng)**
- **Easily automate** with on-magnet washes, fewer cleanup and handling steps, as well as generous overages
- Leverage a **novel reverse transcriptase** tailored specifically for RNA sequencing

APPLICATIONS

- Gene expression analysis
- Gene fusion detection
- Isoform/alternative splicing analysis
- Novel transcript discovery
- Single nucleotide variant (SNV) detection
- Pathway analysis

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ENABLE HIGH-THROUGHPUT OPERATIONS WITH A SIMPLE AND AUTOMATABLE WORKFLOW

Fewer bead cleanups paired with combined and shortened reaction steps deliver sequencing-ready libraries in under 5 hours. Additionally, "on-magnet" bead washing during mRNA capture omits the need to resuspend beads during wash steps, thereby reducing pipetting and magnetting times. Automation is further supported with generous reagent overages to meet dead volume requirements of liquid handlers.

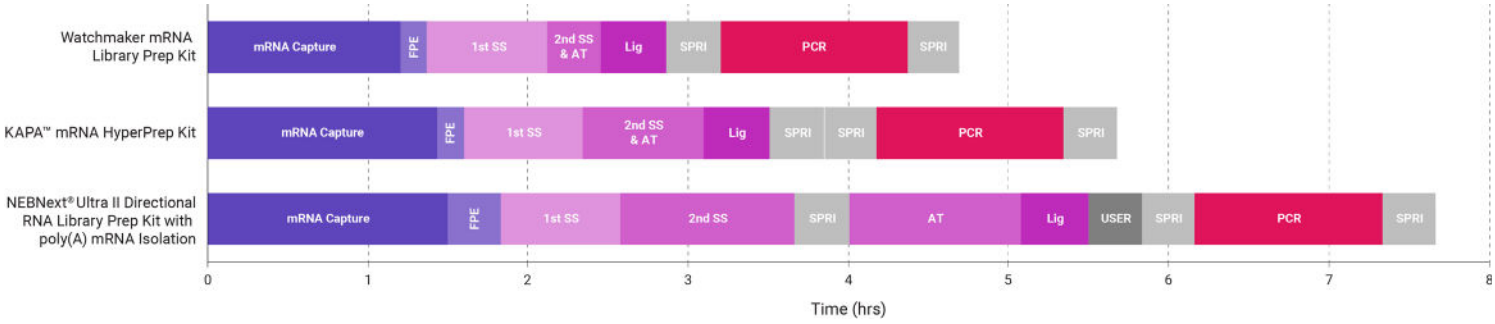

FIGURE 1. Simple, rapid workflow for scalable sample processing. The Watchmaker mRNA Library Prep Kit combines and shortens reaction steps and has fewer cleanups than other commercially available kits.

MAXIMIZE SEQUENCING ECONOMY WITH A HIGH-EFFICIENCY LIBRARY PREP

Uninformative ribosomal RNA (rRNA) and duplicate reads effectively limit the capacity of a sequencing run. Reduce the number of wasted reads with a solution that provides excellent mRNA enrichment and library prep efficiency across a 400-fold input range. This wide input range enables the use of a single workflow without needing specialized low-input solutions that don't maintain strand origin information.

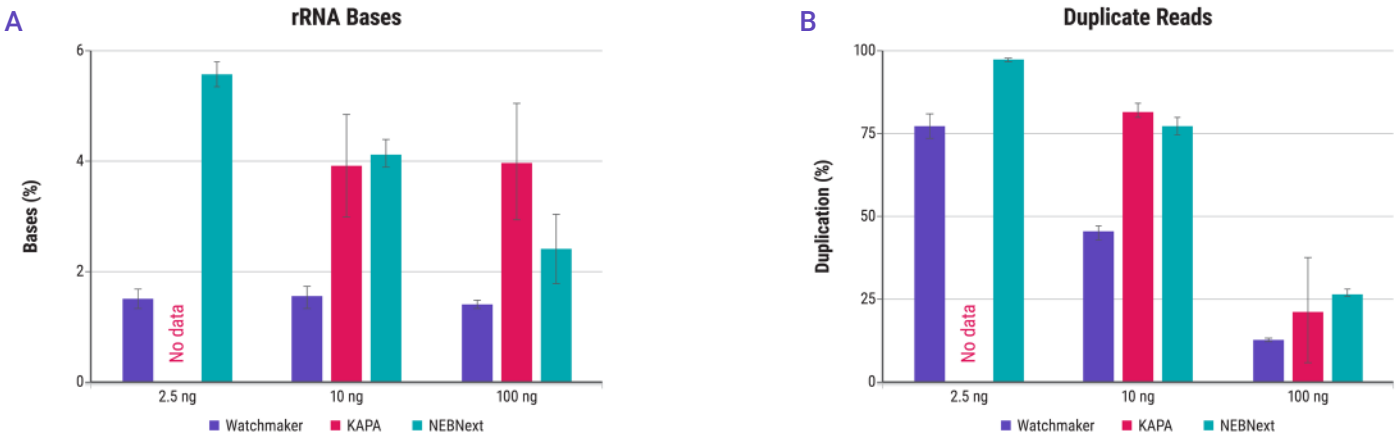

FIGURE 2. Waste fewer bases to rRNA contamination and duplicate reads. Bulk Total RNA extracted from breast tissue (RIN 7) was used to prepare libraries in quadruplicate from a range of RNA mass inputs, as indicated, using the Watchmaker mRNA Library Prep Kit, KAPA mRNA HyperPrep Kit, and the NEBNext Ultra II Directional RNA Library Prep Kit with poly(A) mRNA Magnetic Isolation. Supplier recommendations were used for each workflow. KAPA failed to produce libraries at 2.5 ng. One replicate KAPA library was omitted from analysis due to higher-than-anticipated residual rRNA. Libraries were downsampled to 5M reads. The Watchmaker solution:

(A) results in effective rRNA removal across the full RNA input range, and

(B) delivers improved library prep efficiency which minimizes duplicate reads.

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GET MORE INFORMATION OUT OF CHALLENGING LOW-INPUT SAMPLES

RNA library prep efficiencies plummet with limited sample input. This creates a complexity bottleneck and makes it difficult to derive useful biological insights. The Watchmaker mRNA Library Prep Kit is specifically optimized for improved performance with low inputs and delivers superior library complexity in the form of increased unique gene detection and reduced duplicate reads (see Figure 2).

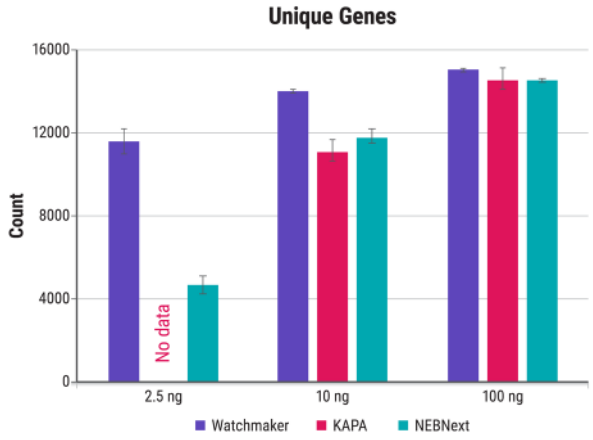

FIGURE 3. Increase library complexity and gene detection sensitivity with low inputs. In addition to a much improved workflow, the Watchmaker solution detects significantly more unique genes with low-input samples and an equivalent number of genes when RNA mass is not limiting. Unique genes were defined as those supported by at least 5 unique raw reads.

See Figure 2 for additional experimental details.

BETTER PRESERVE GENE EXPRESSION DATA INTEGRITY

Gene expression data distortion is another symptom of inefficient library preparation, where many solutions lose relative coverage of lower abundance genes compared to moderate or highly expressed ones. The result is a data output that is not representative of the original sample.

The Watchmaker mRNA Library Prep Kit delivers consistent quantitative expression information across a broad input range to provide confidence in results.

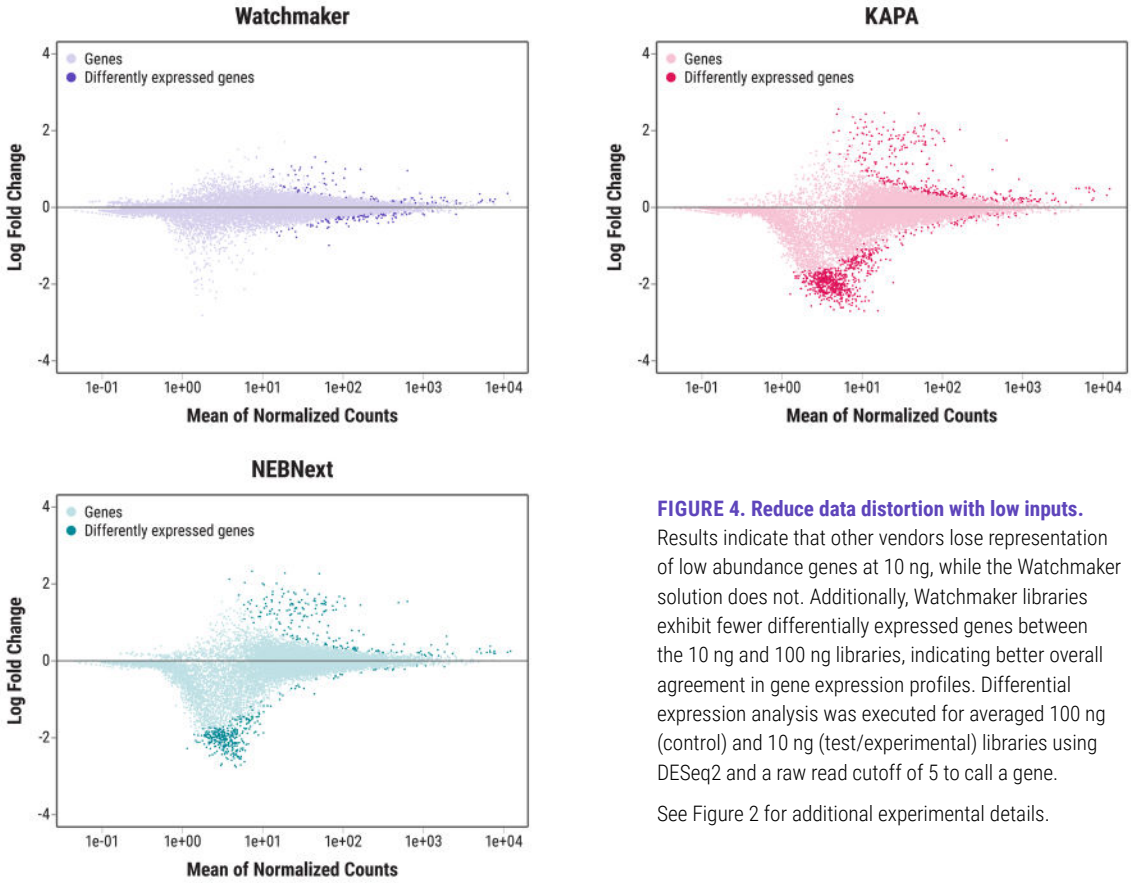

FIGURE 4. Reduce data distortion with low inputs. Results indicate that other vendors lose representation of low abundance genes at 10 ng, while the Watchmaker solution does not. Additionally, Watchmaker libraries exhibit fewer differentially expressed genes between the 10 ng and 100 ng libraries, indicating better overall agreement in gene expression profiles. Differential expression analysis was executed for averaged 100 ng (control) and 10 ng (test/experimental) libraries using DESeq2 and a raw read cutoff of 5 to call a gene.

See Figure 2 for additional experimental details.

IMPROVE BASE COVERAGE — EVEN ACROSS GC-RICH REGIONS

Even transcript coverage uniformity is critical for sensitive applications such as alternative splicing and isoform analysis. The Watchmaker mRNA Library Prep Kit provides consistent coverage uniformity across RNA inputs and leverages Equinox DNA Polymerase to provide improved coverage of GC-rich regions.

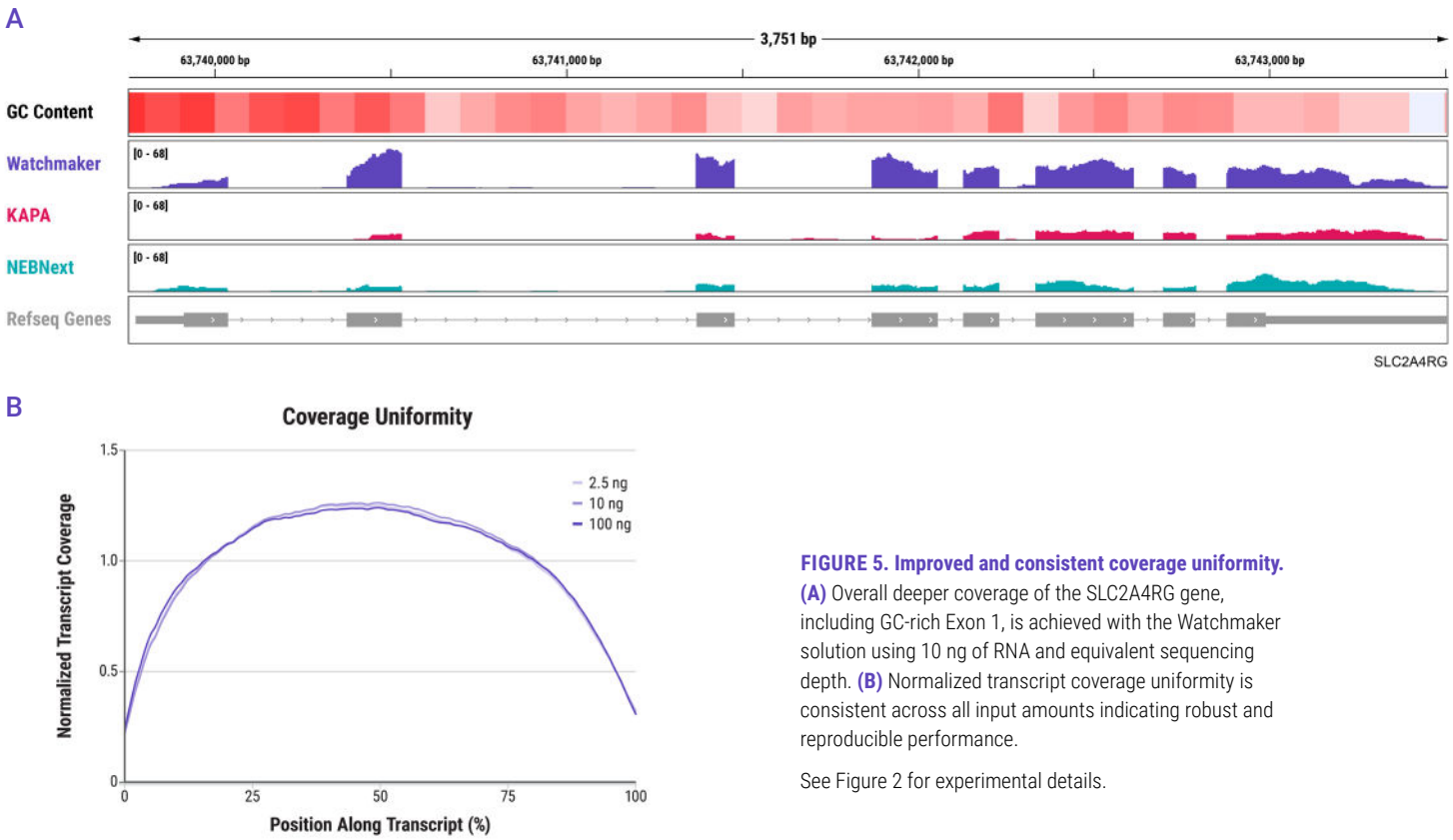

FIGURE 5. Improved and consistent coverage uniformity. **(A)** Overall deeper coverage of the SLC2A4RG gene, including GC-rich Exon 1, is achieved with the Watchmaker solution using 10 ng of RNA and equivalent sequencing depth. **(B)** Normalized transcript coverage uniformity is consistent across all input amounts indicating robust and reproducible performance.

See Figure 2 for experimental details.

Contact sales@watchmakergenomics.com
or visit watchmakergenomics.com/rna-solutions to learn more.

PRODUCT	24 RXN	96 RXN	384 RXN
Watchmaker mRNA Library Prep Kit *includes reagents required for mRNA capture, mRNA library prep, and amplification*	7BK0001-024	7BK0001-096	7BK0001-384

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