Automation of Roche's KAPA EvoPrep Kit for NGS Library Prep on the Beckman Coulter Life Sciences Biomek i7 Hybrid Workstation

Elisa Vega[1], Marsha McMakin[1], Rachel Kasinskas[1], Sean Chien[2], Che Makanjee[2] and Zach Smith[3]
[1]Roche Diagnostics Corporation, Wilmington, MA
[2]Roche Diagnostics Solutions, Pleasanton, CA
[3]Beckman Coulter Life Sciences, Indianapolis, IN

INTRODUCTION

The impact of next-generation sequencing (NGS) on our understanding of human health and disease continues to grow, especially as whole-genome sequencing (WGS) and whole-exome sequencing (WES) become more cost effective. Thus, there is also a growing need for faster, scalable, reproducible workflows. While the automation of NGS library preparation can provide the necessary efficiency and reliability, these workflows must also be compatible with the wide variety of sample types used for these studies–including such challenging inputs as cell-free DNA (cfDNA), DNA from formalin-fixed paraffin-embedded tissue (FFPET), and other sample types with high fragmentation/degradation, chemical modifications, or low concentration.

The **NEW KAPA EvoPrep Kit** streamlines library preparation workflows for various DNA sample types and input concentrations, and **has now been automated on the Beckman Coulter Life Sciences Biomek i7 Hybrid Workstation**.

- The KAPA EvoPrep Kit is effective with mechanically fragmented DNA, cfDNA, and FFPET-derived DNA–using a single workflow.
- The kit's **automation-friendly ReadyMixes** (available in tubes or pre-plated) further improve workflow efficiency and reduce hands-on time while providing reliable and consistent library preparation.

This study demonstrates the successful automation of the **KAPA EvoPrep DNA Library Prep** workflow on the **Biomek i7 Hybrid Workstation**, and provides a new automated method for high-throughput library construction with challenging sample inputs, enabling users to maximize their library prep efficiency.

AUTOMATED KAPA EVOPREP WORKFLOW

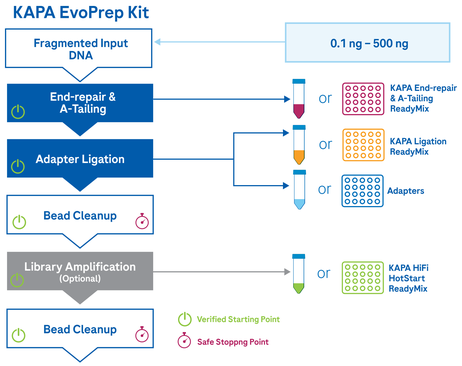

Figure 1: Automated KAPA EvoPrep Workflow. On the left the diagram shows the automated KAPA EvoPrep workflow with full-length KAPA UDI Adapters. The run time is estimated based on a 96-sample run with Plated ReadyMix reagents. The method offers the flexibility to start at any step indicated by the green "power button." The red timer indicates the safe stopping points in the workflow per the instructions for use. On the right the diagram shows the available reagent formats that can be used with the automated method (tubed or pre-plated). *Note: For inputs greater than 50 ng (up to 500 ng) processed with full-length KAPA UDI Adapters follow a workflow that is PCR-Free; all supported inputs processed with KAPA Universal Adapter and KAPA UDI Primer Mixes require library amplification. Cycle number recommendations are provided in the instructions for use.*

METHODS

Library Preparation on the Beckman Coulter Life Sciences Biomek i7 Hybrid Workstation

- **Human genomic DNA (gDNA) was mechanically sheared** to a target fragment size of ~250 bp on a Covaris M220 instrument.
- **High-throughput processing capability and edge effects** were assessed using high-throughput runs with 10 ng of input DNA. Two 96-sample runs were performed on the Biomek i7 Hybrid Workstation: (1) using full-length KAPA UDI Adapters with tubed ReadyMix reagents, and (2) using KAPA Universal Adapter* and KAPA UDI Primer Mixes with pre-plated ReadyMix reagents. Equivalent libraries were also created manually (n=4 per workflow).
- **Performance across a range of input amounts** was assessed by using various amounts of sheared gDNA as input into library prep in the following amounts: 0.1 ng, 1.0 ng, 10 ng, 100 ng, and 500 ng. Libraries were generated on the Biomek i7 Hybrid Workstation on a low-throughput run using the KAPA EvoPrep Kit (n=4 per input) with full-length KAPA UDI Adapters. Equivalent libraries were also generated manually (n=4).
- **Site-to-site performance** on the Biomek i7 Hybrid Workstation was also assessed by performing an equivalent high-throughput run at two different Roche sites using 10 ng DNA with full-length KAPA UDI Adapters (using tubed reagents).

Assessment of total yield and fragment size:

- Total yield and fragment size metrics of both automated and manual libraries were assessed for downstream processing capability. Libraries were assessed using the Qubit 4.0 with the Qubit 1X dsDNA HS (high-sensitivity) assay kit and the 2100 BioAnalyzer instrument with the High Sensitivity DNA Kit.

*KAPA Universal Adapter is only available in tubed format.

RESULTS

High-throughput Library Preparation with KAPA EvoPrep Kit Tubes

(Tubed ReadyMix Reagents and Full-length KAPA UDI Adapters)

Figure 2: High-throughput automated library preparation yielded equivalent results without introducing edge effects, using tubed Ready Mix reagents and full-length KAPA UDI Adapters. Libraries were prepared from 10 ng sample input of mechanically sheared NA12878 human gDNA on the Biomek i7 Hybrid Workstation, as well as manually where indicated. **(A)** Per-well yields for both interior and exterior well positions, demonstrating comparable performance, minimal variation, and a CV of 6.6%. Well A01 in the 96 well plate is designated as position 1 and well H12 as position 96; this order is followed in columnar fashion (down each column and to the top of the next column etc). **(B)** The average total library yield from automated and manual library preparation, demonstrating improved yields with automation. **(C)** Fragment size distribution for libraries generated on the Biomek i7 Hybrid Workstation and manually, demonstrating consistent sizing.

High-throughput Library Preparation with KAPA EvoPrep Kit Plates

(Pre-Plated ReadyMix reagents and KAPA Universal Adapter* with KAPA UDI Primer Mixes)

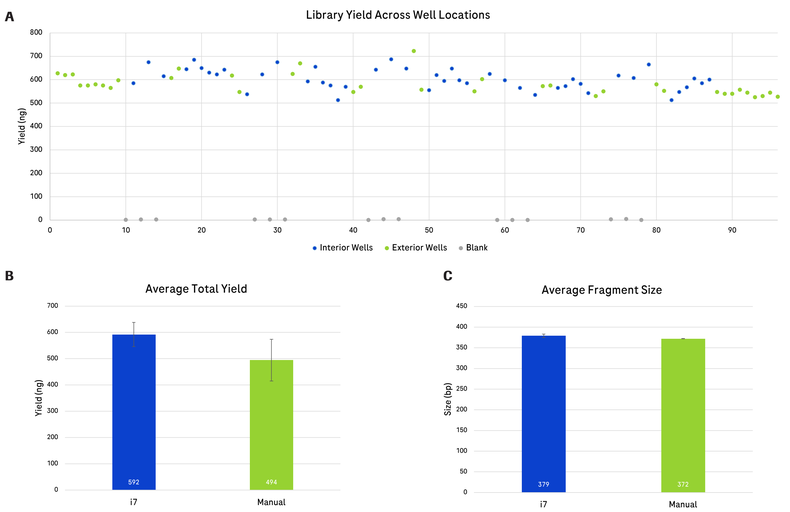

Figure 3: High-throughput automated library preparation yielded equivalent (or better) results without introducing edge effects, using plated ReadyMix reagents and KAPA Universal Adapter* with KAPA UDI Primer Mixes. Libraries were prepared from 10 ng sample input of mechanically sheared NA12878 human gDNA on the Biomek i7 Hybrid Workstation, as well as manually where indicated. **(A)** Per-well yields for both interior and exterior well positions, demonstrating comparable performance, minimal variation, and a CV of 7.7%. Well A01 in the 96 well plate is designated as position 1 and well H12 as position 96; this order is followed in columnar fashion (down each column and to the top of the next column etc). **(B)** The average total library yield for automated and manual library preparation, demonstrating improved yields with automation. **(C)** Fragment size distribution for libraries generated on the Biomek i7 Hybrid Workstation and manually, demonstrating consistent sizing.

Automated Library Preparation with Various Sample Input Amounts

(Tubed ReadyMix Reagents and Full-length KAPA UDI Adapters)

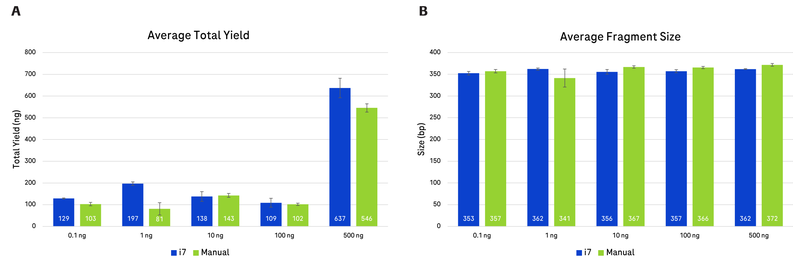

Figure 4: Automation of KAPA EvoPrep Kit across a 5000-fold range of input amounts resulted in equivalent-or-better yields and comparable fragment sizes. Libraries were generated from 0.1 ng, 1 ng, 10 ng, 100 ng and 500 ng sample inputs of mechanically sheared NA12878 human gDNA on the Biomek i7 Hybrid Workstation and manually. Graph **(A)** shows library yields for each input amount. Graph **(B)** shows the comparison of the average fragment sizes. A comparison of the automated libraries and the manual libraries demonstrates that the automated method generates libraries with improved yields over that of manual preparation while maintaining consistent library sizes.

Cross-site Comparison of Automated Library Prep using KAPA EvoPrep Kit on the Biomek i7 Hybrid Workstation

(Tubed ReadyMix Reagents and full-length KAPA UDI Adapters)

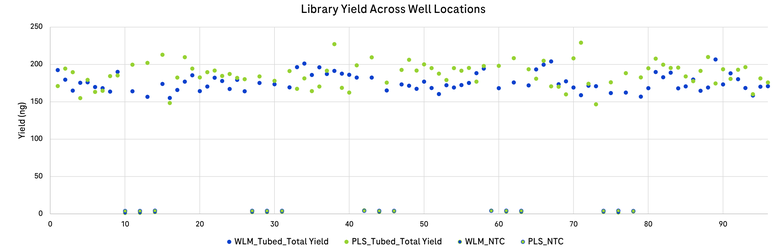

Figure 5: Consistent yield was achieved across multiple sites using two different Biomek i7 Hybrid Workstations and KAPA EvoPrep Kit with Full-Length KAPA UDI Adapters. Libraries were created from 10 ng sample input of mechanically sheared NA12878 human gDNA on two Biomek i7 Hybrid Workstations located at Roche laboratories in Wilmington, MA. [WLM] and Pleasanton, CA. [PLS]. Comparison of the yield of all libraries demonstrates comparable performance and minimal variation with a CV of 6.6% for WLM and 8.5% for PLS.

CONCLUSIONS

Automation of Roche's new KAPA EvoPrep Kit on the Beckman Coulter Life Sciences Biomek i7 Hybrid Workstation produces NGS libraries with consistent fragment size distribution and sufficient yield for sequencing across a 5000-fold range of input amounts.

The library quality metrics demonstrate that these automated high- and low-throughput methods:

- Generate NGS libraries with consistent yields comparable to manual processing, without edge effects.
- Deliver consistent yields and fragment sizes across a wide range of input amounts.
- Provide consistent results across sites, users, and Biomek i7 Hybrid Workstation instruments.

Overall, **the automation of Roche's new KAPA EvoPrep Kit on the Biomek i7 Hybrid Workstation** offers a solution that improves reproducibility while providing high-throughput capability for challenging sample inputs in DNA library preparation.

Explore support for non-Roche liquid handlers

Learn about KAPA EvoPrep Kits

Data on file at Roche
Project: Biomek i7 EvoPrep
For Research Use Only. Not for use in diagnostic procedures.
© 2025 Roche Sequencing & Life Science. KAPA is a trademark of Roche. All other trademarks are property of their respective owners.
© 2025 Beckman Coulter, Inc. All rights reserved. Beckman Coulter, the stylized logo, and the Beckman Coulter product and service marks mentioned here in are trademarks or registered trademarks of Beckman Coulter, Inc. in the United States and other countries. All other trademarks are the property of their respective owners. 2024-GBL-EN-107098-v2 (v2.1)

MC-US-16337 A850 01/25