



Automated Workflow Overview

Watchmaker Genomics and PerkinElmer have collaborated to automate the Watchmaker® DNA Library Prep Kit with Fragmentation on the Sciclone® G3 NGSx workstation, offering a high throughput workflow for DNA library prep with enzymatic fragmentation that reduces both the chance for human error and overall hands-on time. The automated workflow on the Sciclone G3 NGSx workstation can process up to 96-samples in a single run and includes a flexible user interface that accommodates a variety of workflow options supported by the Watchmaker kit.

Automated Workflow Features

- Reagent Workbook: Details master mix formulations, volumes, plate types and relevant deck locations of all reagent containing consumables required to run
- Push Button Protocols: Allows users to choose between PCR
 v. PCR-free workflows at the start of the run
- Flexible User Interface: Supports 1 96 sample processing, stubby v. full-length adapters and dynamic bead cleanup ratios (Figure 2)
- **Deck Images:** Guides users through the placement of labware on-deck with simple step-by-step instructions (Figure 3)

DNA input samples prepared in 96-well plate

Broadcast the 5X Frag/AT master mix and add to samples (15 min)

Frag/AT incubation off-deck (35 - 55 min)

Broadcast Adapter Ligation master mix (15 min)

Add Adapters and Ligation master mix to samples (5 min)

Ligation incubation on-deck at 20°C (15 min)

Post-Ligation Cleanup (30 min)

Broadcast PCR mastermix and add to samples (15 min)

Library Amplification off-deck (15 - 60 min)

Post-Amplification Cleanup (30 min)

Figure 1: Automated process for Watchmaker® DNA Library Prep Kit with Fragmentation on the Sciclone G3 NGSx workstation. Total processing time for 96 samples, including reagent and deck setup, is between 3.5 – 4.5 hours depending on the Frag/AT and PCR parameters. All automated steps are colored purple and manual interventions are colored pink.



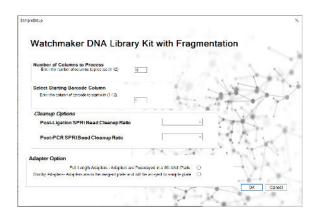


Figure 2: Flexible User Interface

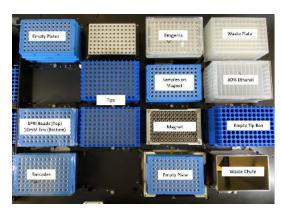
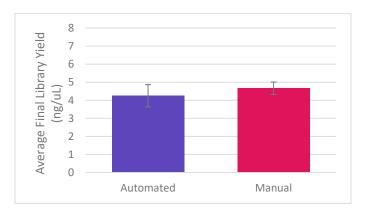


Figure 3: Deck layout of the Sciclone G3 NGSx showing placement of reagents and consumables

Automated Workflow Performance

To initially assess the performance of the automated workflow, 8 manual and 96 automated samples were prepared using 10 ng of total Promega Human Genomic DNA. Workflow conditions were selected based on the Watchmaker DNA Library Prep Kit with Fragmentation User Guide and included a 20 min fragmentation at 37°C and 4 cycles of PCR. 48 No Template Controls (NTCs) were included in the automated run to check for cross-contamination. Final library yields and sizes were measured using the Qubit dsDNA High Sensitivity assay (Figure 4) and LabChip® NGS 3K Assay (Figure 5). Initial QC results were comparable between manual and automated libraries, with no observed cross-contamination in NTCs. The automated workflow also resulted in no obvious plate effects. Both final library yields and size distribution profiles were highly reproducible even across a 96-well plate. Additional testing using more challenging sample types will be performed with sequencing metrics to further assess the performance of the automated workflow.



 $\textbf{\textit{Figure 4:}} \ \textit{Final library yields and sizes were measured using the Qubit dsDNA \textit{High Sensitivity assay}}$

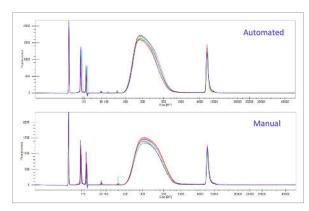


Figure 5: Final library yields and sizes were measured using the LabChip® NGS 3K Assav

The Watchmaker® DNA Library Prep Kit with Fragmentation automated solution for the PerkinElmer Sciclone G3 NGSx workstation will be available once final performance testing is complete. Please contact your local Watchmaker or PerkinElmer representatives for more information.

Contact Information

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