



ACCEL-NGS® XL LIBRARY KIT FOR PACIFIC BIOSCIENCES®

Generate Quality Genome Assemblies. Faster.

Highlights

- **Faster and easier protocol**
Process your samples with a simple workflow and have sequence-ready libraries in 1 day.
- **Longer average read length**
Achieve maximum average read length from DNA fragments sheared to ≥ 20 kb.
- **Less input DNA**
Work with challenging or limited sample quantities as little as 2 μg .



Introduction

Long read sequencing technology provides better coverage and assembly metrics for applications like *de novo* assembly and whole-genome sequencing. Despite these benefits, the current long-read SMRTbell™ template preparation protocol is labor intensive and requires high sample inputs, while offering low library conversion efficiency.

To meet these challenges, Swift Biosciences developed the AcCEL-NGS XL Library Kit for long read sequencing from DNA fragments sheared to ≥ 20 kb. The core of this kit is based on AcCEL-NGS 2S technology leveraging Swift's enhanced repair chemistry and adapter dimer-free ligation enzymology. This produces the longest possible contiguous polymerase reads and increases library conversion efficiency from low input quantities. Optimal performance is achieved with high quality DNA samples and the BluePippin™ DNA Size Selection System.

In addition to components necessary for generating libraries, the kit includes final repair reagents, a sequencing primer, and a 10X primer buffer to perform the subsequent repair and annealing steps. This easy protocol provides a fast workflow compatible with PacBio sequencing technology.

Faster, Simpler Workflow

This simple workflow facilitates more gentle handling to reduce DNA damage by less pipetting, shorter incubations, and no heat steps or exonuclease digestion steps. Using five simple incubations, this protocol repairs both 5' and 3' termini and attaches adapter sequences to the ends of fragmented double-stranded DNA, while preserving DNA integrity.

The Accel-NGS XL workflow requires as little as 2 µg of DNA input (sheared to 20 kb) per sample and is compatible with RS II platforms.

Workflow Comparison

The Accel-NGS XL workflow is a fast, efficient workflow consisting of four repair steps spanning 2.5 hours, one 30-minute ligation, and five bead DNA purification steps. The total protocol is approximately 1 day, from shearing to sequence-ready library. It enables repair and ligation of long templates using a 'with-bead'-based purification, in which beads are added to the first clean-up step and are retained and recycled throughout subsequent enzymatic reactions and clean-up steps without the need for sample tube transfers. By integrating these activities into a simple format, it minimizes hands-on time, sample loss, and processing errors.

dsDNA Fragment

Repair I

End Repair

⌚ 10 minutes

Repair II

Internal Repair

⌚ 60 minutes

Repair III

Polishing

⌚ 20 minutes

Ligation

Ligation of Adapter

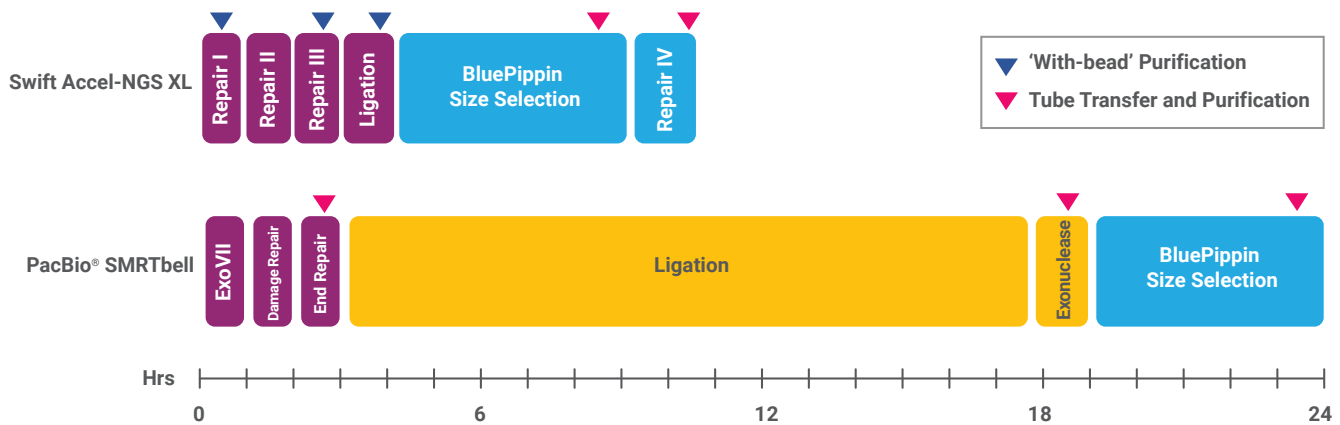
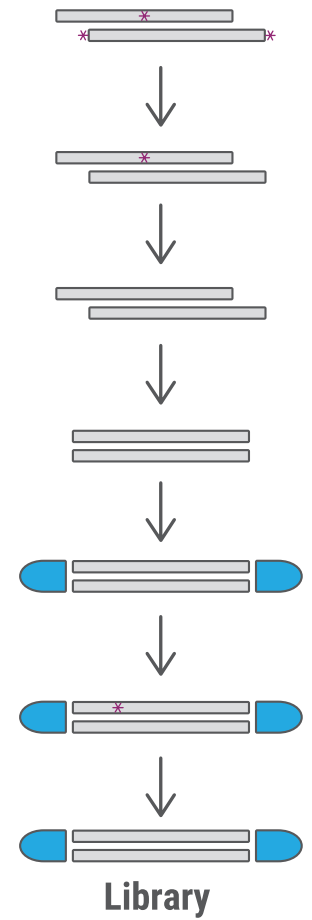
⌚ 30 minutes

BluePippin Size Selection

Repair IV

Internal Repair

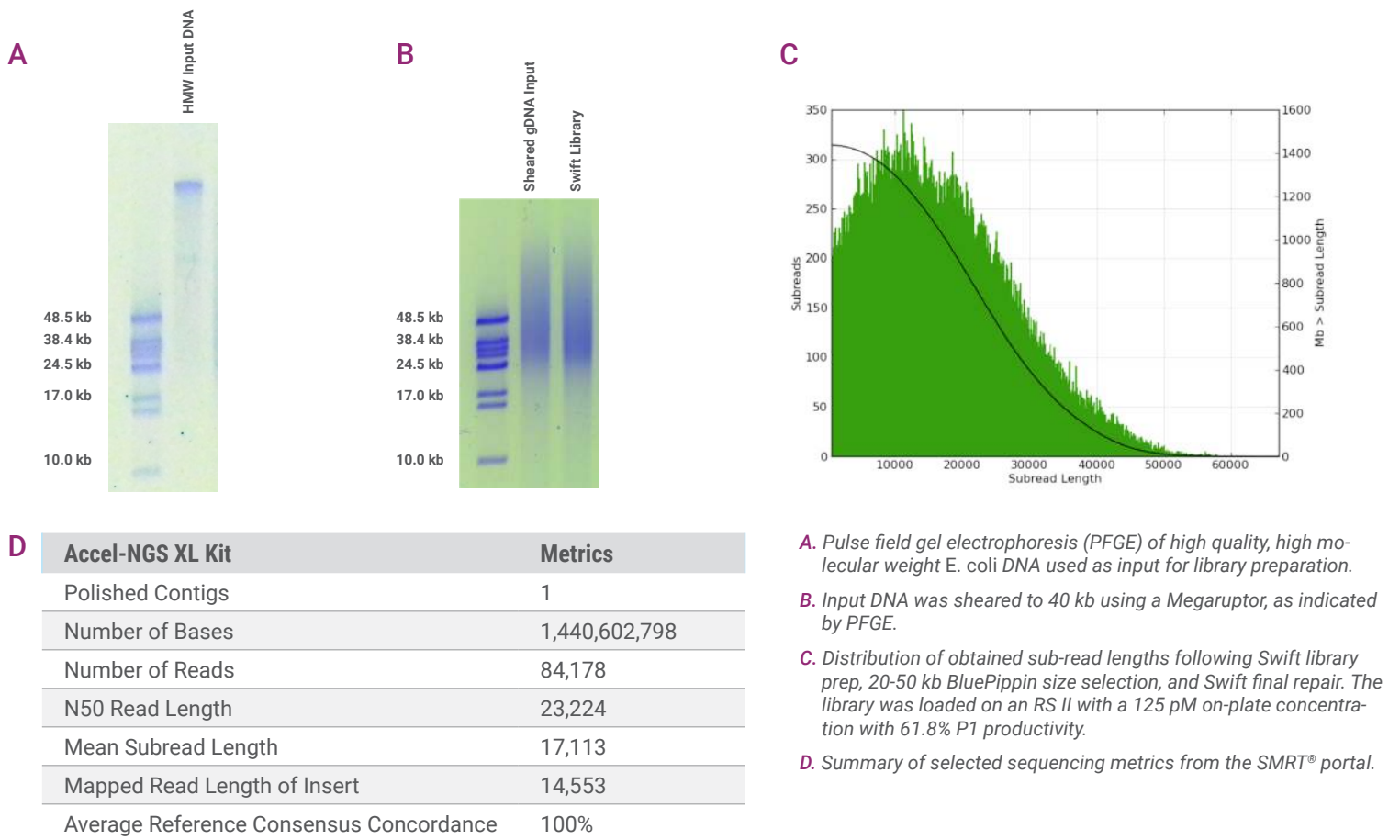
⌚ 60 minutes



This diagram shows Swift's workflow with five bead clean-up steps as compared to the PacBio workflow with overnight ligation and multiple tube transfers. Each color indicates a tube transfer. When comparing workflow diagrams, refer to Pacific Biosciences' Procedure and Checklist - 20 kb Template Preparation Using BluePippin Size-Selection System.

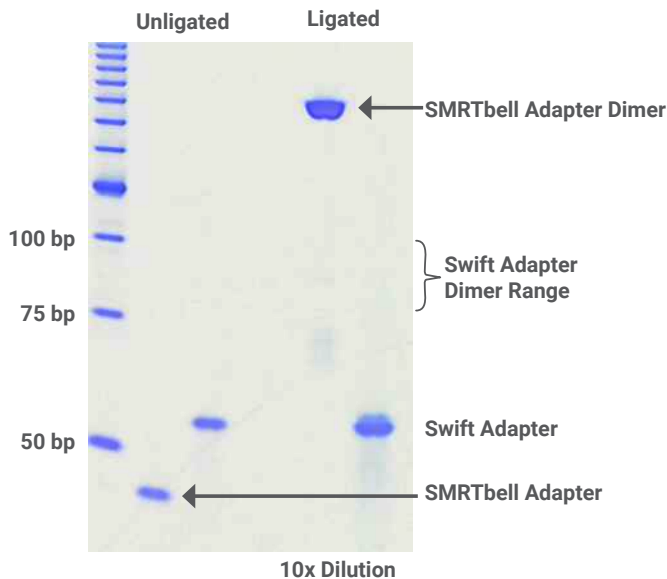
Maximized Average Read Length

Accel-NGS XL Library Kit Provides Long Average Read Length



Accel-NGS XL Library Kit Offers Adapter Dimer-Free Ligation Chemistry

This kit offers a fast, efficient ligation step that prevents adapter dimers. Based on Swift's proprietary 2S technology, the protocol eliminates laborious post-library prep clean-up steps, thereby allowing use of the BluePippin size selection prior to library preparation, if desired.



A ligation reaction was performed as recommended for a 20 kb DNA shear in the absence of DNA substrate. This enabled identification of adapter dimers that form during ligation. For the SMRTbell ligation reaction, the ExoIII/VII purification was performed to remove any non-covalently closed molecules. Each sample was diluted from the final eluate as indicated and ran on a 15% Urea-PAGE gel. The SMRTbell adapter dimers migrate abnormally due to secondary structure. Lane 1: Ladder. Lanes 2-3: Unligated adapter control. Lanes 4-5: Each adapter plus ligase.

Specifications

| Specification | Feature | Accel-NGS XL Library Kit |
|---------------|-----------------------|--|
| Input | Input DNA required | 2 µg or greater |
| | Sample types | Microbial, plant, animal, and human |
| | Sample quality | High molecular weight DNA > 50 kb |
| Workflow | Assay time | Hands-on time: 5 hours Size selection time: 2-8 hours Note: The average size selection time is 5 hours, assuming a 20-50 kb size selection. |
| | System compatibility | PacBio RS II Sequel™ (in development) |
| Design | Kit size | 16 libraries per kit |
| | Fragmentation method | Covaris® g-TUBE, Diagenode® Megaruptor® |
| | Fragment size | ≥ 20 kb sheared |
| | Final repair reagents | Included, standard to kit |
| Performance* | Mean subread length | 17,113 |
| | N50 | 23,224 |
| | Conversion efficiency | 85% |

* Performance specifications are based on E. Coli genomic DNA prepared from a 40 kb Megaruptor shear. 20-50 kb fragments were selected using the Sage BluePippin.

Ordering Information

| Product Name | Reactions | Catalog No. |
|---|-----------|-------------|
| Accel-NGS XL Library Kit Includes library reagents, sequencing buffer, and 10X annealing buffer. All components stored together at -20 °C. | 16 | 71016 |

Visit www.swiftbiosci.com for easy ordering.



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