Application Note

Automated Library Preparation from 0.5 ng of ChIP DNA

Apollo 324™ System

Data contributed by Dr. Michael Quail of the Wellcome Trust Sanger Institute.

Overview

This application note describes Illumina® library preparation from 0.5 ng of chromatin immunoprecipitation (ChIP) sample DNA input with the Apollo 324™ System at the Wellcome Trust Sanger Institute.

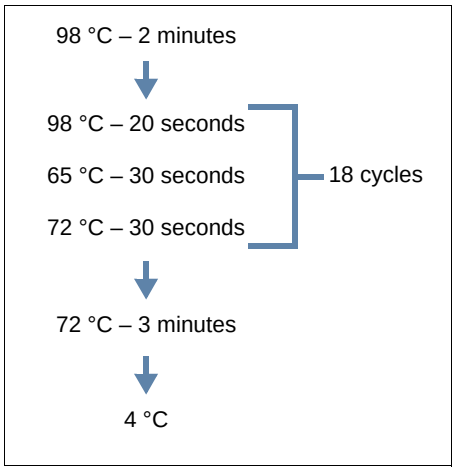
ChIP sample preparations generally yield low amounts of DNA, sometimes less than 1 ng. The Apollo 324 System automates library preparation for Illumina next-generation sequencing using genomic DNA, cDNA and ChIP sample input. In this experiment, an Illumina library preparation was performed using an input of 0.5 ng total DNA. This input concentration mimics the low amounts of DNA expected from ChIP input samples.

Method

Conditions for post-PCR processing were optimized to prepare DNA libraries from low-input CHiP samples. In this experiment, 15 µL containing 0.5 ng of sheared *Salmonella pullorum* genomic DNA was run on the Apollo 324 using the PrepX™ DNA Library protocol. The adapter concentration used in the PrepX DNA Library ligation recipe was 1.7µM of paired-end adapter. After the automated library preparation, a portion of the resulting sample was added to the following PCR recipe:

Component	Volume
DNA	5 µL
Index Primer	1 µL
PE1.0	1 µL
Water	18 µL
2x PCR Premix	25 µL
Total Volume	**50 µL**

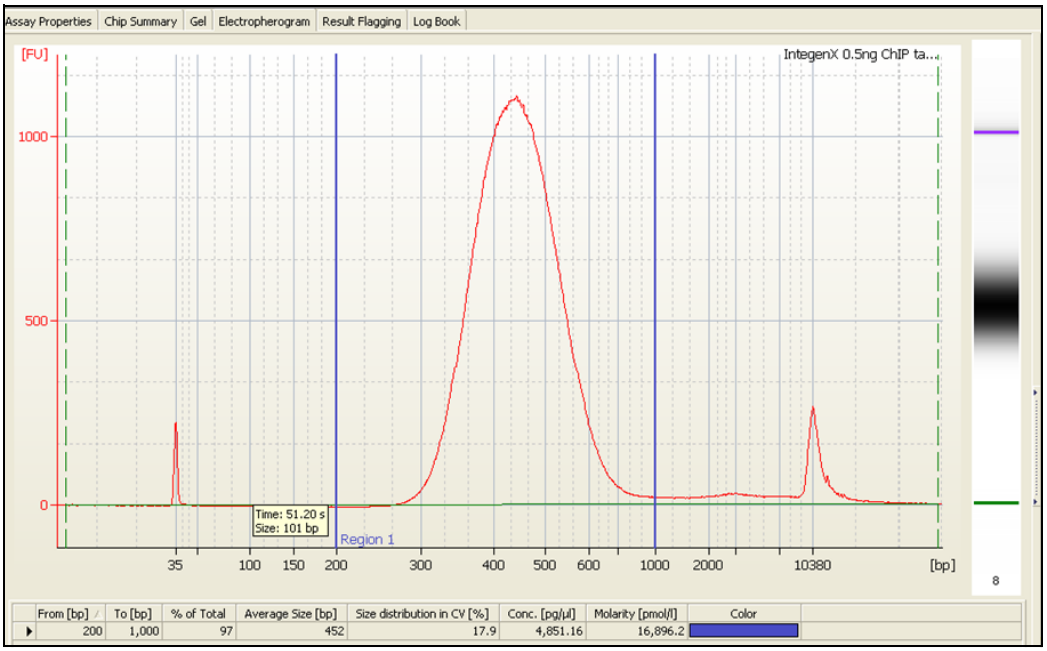
This sample PCR mix was then subjected to 18 cycles of PCR, using the following steps:



The PCR sample was then cleaned up by adding 60 µL of AMPure® XP in 15.6% PEG. After EtOH washing, the captured sample was eluted into 50 µL of water.

Results

Very low input samples require optimization of post-PCR conditions to obtain sufficient and meaningful results. Starting with a sample containing only 0.5 ng of DNA, and using the automated Apollo 324 library preparation system, over 200 ng of DNA ready for Illumina next-gen sequencing was recovered after PCR. The Apollo 324 System can create high quality DNA libraries from low yield samples resulting from ChIP sample preparations.



0.5 ng of sheared genomic DNA input on the Apollo 324, after 18 cycles of PCR

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