Introduction

The reduction in costs and speed of sequencing has fueled the expansion of next generation sequencing (NGS). However, there are still barriers preventing wider adoption of the technology. One of them is the lack of a robust, easy-to-use workflow for low throughput library preparation to minimize human error and the time spent on tedious manual pipetting. The BioQule™ NGS System is a complete solution combining automation, reagents, consumables, and scripts, enabling walkaway automation and quantitation for low-throughput library prep. This low-cost, benchtop instrument delivers libraries ready to load into your sequencer in only 15 minutes of hands-on time (HOT). It can automate most library prep workflows. In this note we focus on the results obtained when using the BioQule™ NGS System for Whole Genome Sequencing (WGS) with enzymatic fragmentation.
Methods

Forty WGS libraries were constructed using the NEXTFLEX® Rapid XP V2 DNA-Seq kit (NOVA-5149-503) with NEXTFLEX® Unique Dual Index Barcodes (NOVA-514150) on the BioQule™ NGS System. Five runs of 8 samples each were prepared using 40 ng of 24 Human Promega® DNA as the starting material.

Libraries were quantified in the BioQule™ NGS System and their size was analyzed using a High Sensitivity assay. Following this, libraries were pooled and run on an Illumina® MiniSeq® platform at 2x150.

Results

The final volume of library obtained was 28 µL, with an average yield of 13.29 ng/µL (Figure 1). The libraries aligned with the expected range with target peak ~ 425 bp. The % dimer was only 0.032±0.019%.

![Figure 1. Highly reproducible yields obtained using the BioQule™ NGS System. Quantitation was performed automatically on the BioQule™ NGS System after library preparation was completed.](image)

The sequencing metrics for these WGS libraries can be seen in Figures 2-3

![Figure 2. Insert size for 40 the libraries prepared on the BioQule™ NGS System, grouped by run.](image)

![Figure 3. Analysis of GC bias from the 40 libraries obtained with NEXTFLEX® Rapid XP V2 DNA-Seq kit. Results show minimal variability in normalized coverage across 20-60% GC, encompassing ~ 95% of the human genome (grey bars).](image)

Conclusion

The NEXTFLEX® Rapid XP V2 DNA-Seq kit automated on the BioQule™ NGS System produced consistent libraries ready to be sequenced on an Illumina® sequencing platform. The yield and size of libraries were within the expected range. The BioQule NGS workflow allows the creation of up to 8 libraries with only 15 minutes of hands-on user time in 4 easy steps, making NGS accessible to labs of all sizes and technical experience.