Utilizing unique dual indexes (UDIs) may be advantageous for sensitive genomics applications or to counteract the effects of index hopping on patterned flow cell sequencers. However, most available UDI methods incorporate indexes using individual sample PCR, which can be labor and cost intensive particularly at high scale.

In the purePlex™ DNA Library Preparation workflow, UDIs are added early in the library prep process via sequential Tn5 transposition with full-length UDI adapters. This approach allows sample pooling immediately following these tagging steps. All subsequent purification, amplification, and QC steps are performed on pooled libraries, greatly reducing time and cost.

In addition, tagging is performed in the presence of our novel Normalization Reagent, delivering low CV of read counts with pooled libraries across a 10-fold input range of genomic DNA, cDNA, amplicons, or plasmids.

purePlex™ Workflow

Introduction

**Introduction**

**Results**

**Auto-Normalization**

Samples with varying DNA inputs between 3-30 ng were processed with purePlex Library prep with (+) and without (−) Normalization Reagent. Inclusion of Normalization Reagent leads to a very low read count CV within the pooled library independent of sample input amount.

**Robust performance for all GC contents**

Libraries were prepared from bacterial genomic DNA (ATCC) ranging in GC content from 29-69% using both purePlex and Nextera XT and sequenced on NextSeq 550.

GC bias plots, right, demonstrate purePlex has more even coverage across high and low GC regions.

**Insert Size**

For purePlex, insert size within a pool of samples is consistent regardless of input (panel A) or GC content (panel B). In contrast, Nextera XT insert size varies across different GC contents (panel C).

**Conclusions**

- The purePlex DNA Library Preparation kit normalizes read count and insert size over a 10-fold input range, alleviating the burden of individual sample normalization prior to sequencing.
- Outperforms Nextera XT in terms of GC bias and generates consistent and tunable insert size regardless of sample input and GC content.