Comparison of NuQuant® NGS Library Quantification with other library quantification methods

Genomics

Karen Brown (Karen.Brown@tecan.com), Tecan Genomics, **Lela Andrews**, Tecan Genomics, **Bin Li**, Tecan Genomics, **Lin Pham**, Tecan Genomics

Optimized cluster generation is critical to the success of sequencing runs on Illumina sequencing platforms, and cluster density is directly driven by the amount of library DNA loaded onto the flow cell. For years, the industry standard for library quantification has been qPCR – the only method with sufficient accuracy to ensure optimal flow cell loading. However, qPCR is a time-consuming process requiring multiple processing steps. NuQuant NGS Library Quantification is a proven, proprietary method for measuring the molar concentration of NGS libraries by utilizing fluorescence technology to count functional library molecules irrespective of insert size in under 10 minutes. With NuQuant, a specific number of fluorescent labels are incorporated into the library molecules during library preparation. Direct measurement of these sequenceable molecules gives greater precision in how library pools are diluted to maximize normalization. To determine the accuracy and precision of NuQuant, we compared molarity measured by NuQuant with molarity determined by sequencing on iSeq 100, and found signification correlation between the two methods. Then, we examined qPCR and Qubit, standard methods of library quantification, and how their performance compares to NuQuant in benchmarking studies performed with Celero™ DNA-seq libraries and Universal Plus™ mRNA-seq libraries. We assessed both library concentration and the proportion of reads and found NuQuant and qPCR results to be comparable and functionally equivalent, while Qubit showed lower measurements of library concentration and high variability compared to NuQuant and qPCR. In summary, NuQuant is a simple library quantification method with equivalent accuracy to qPCR and iSeq 100.