



SWIFT NORMALASE KIT

Revolutionary Library Normalization Technology for NGS Laboratories

Highlights

- Saves time and increases throughput**
 Uniform sample processing with fewer handling steps to generate balanced library representation in multiplexed sequencing
- Reduces variability to save on sequencing costs**
 Better balanced libraries allows higher multiplexing per run and fewer sample failures
- Flexible design for many workflows**
 Compatible with diverse library preparation methods and sample types to produce more evenly balanced sequence data

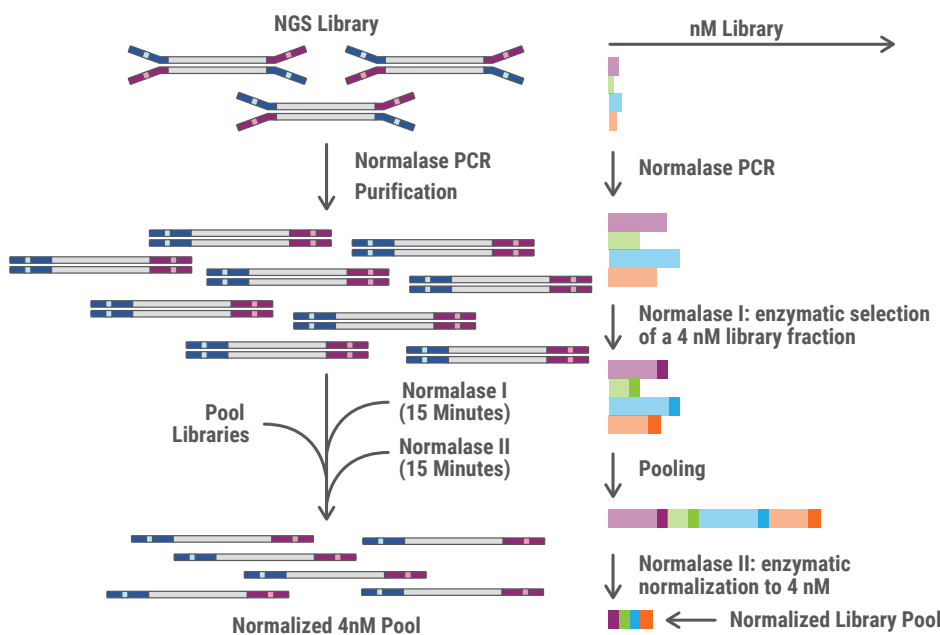


Figure 1 (Left): Conventional library amplification with Normalase primers generates libraries with indexed adapters to $\geq 12nM$, the minimum threshold required to condition the libraries for Normalase I and II incubations. Following a standard purification, 4 nM of each library is enzymatically selected during the Normalase I step. After Normalase I, libraries are combined into a single pool. The Normalase II step enzymatically normalizes all libraries within the pool to 4 nM. Without the need for further purification, the pool is ready for sequencing.

The Swift Normalase Kit offers a novel enzymatic library normalization technology that is augmented by the power to consolidate library normalization and pooling for loading on Illumina® sequencing platforms. The Normalase workflow eliminates the need for library quantification and concentration adjustment prior to library pooling, resulting in optimal cluster density and library balance. The Swift normalization method can easily be integrated into standard protocols to improve turnaround time loading accuracy for NGS laboratories.

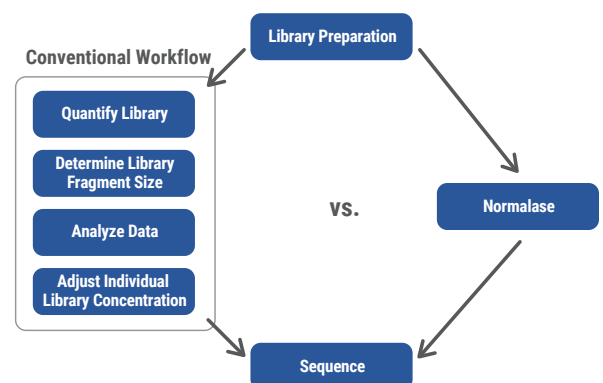


Figure 2 (Above): The first step of Normalase replaces conventional library amplification, but integrates special primers to pre-condition the libraries, which is followed by two 15 minute incubations to normalize libraries to 4 nM within a single pool.

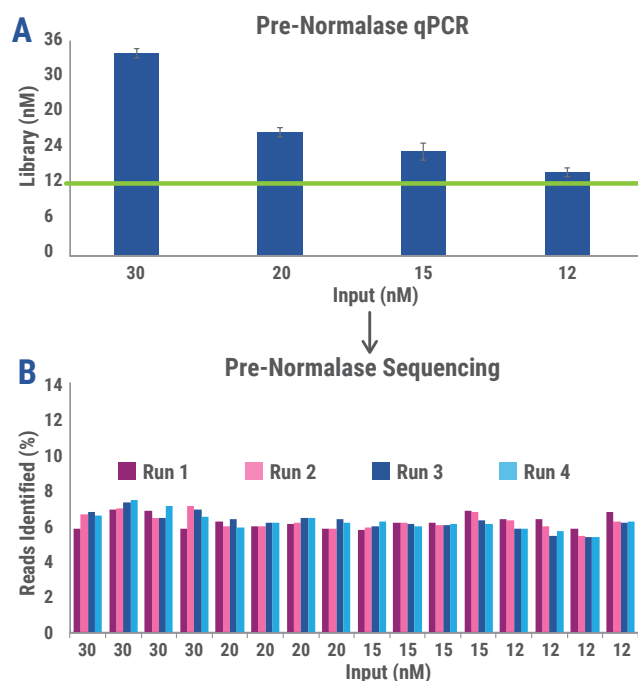
Fastest, Easiest Workflow

The benefits of Normalase to traditional library quantification methods are that normalization is by molarity vs. by mass, is quantification-free, and does not require individual concentration adjustment or fragment size estimate of each library.

High Quality Results

Swift Normalase offers highly reproducible DNA library normalization and robust performance across variable insert sizes.

	Normalase	Qubit	qPCR	BioAnalyzer
Molar Normalization	✓	✗	✗	✗
Quantification-Free	✓	✗	✗	✗
Concentration Adjustment-Free	✓	✗	✗	✗
Size Correction-Free	✓	✗	✗	✗



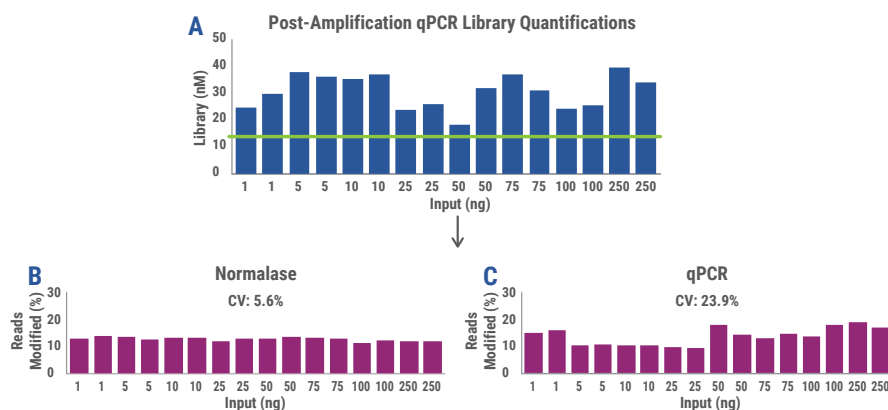
Loading (pM)	Cluster Density (K/mm ²)	# of Libraries	Library Balance (CV%)	Insert Size
12	1370	6	9.7	150
12	804	16	6.5	200
12	918	16	7.1	200
12	972	16	8.0	200
12	1043	16	8.2	200
12	856	16	9.5	350
12	1157	6	5.4	350
12	1070	5	3.7	600

Figure 3 (Left): 16 libraries amplified with Normalase primers to 30, 20, 15, and 12 nM ($n=4$ library/concentration) were used for input into Normalase and processed four independent times. **A:** Libraries were confirmed to meet the minimum threshold at or above 12 nM. Error bars: SD. **B:** Post-Normalase sequencing results for indexing balance of the four independent runs through Normalase using the same 16 libraries. The overall balance variation was calculated to be a coefficient variation (CV) of 7.3%, demonstrating reproducible normalization across Normalase and sequencing runs.

Table 1 (Above): Expected and consistent cluster density generation using MiSeq[®] v2 chemistry at 12 pM from library pools normalized to 4 nM using Normalase.

Robust Library Balancing

Figure 4 (Right): **A:** 16 Swift 2S Turbo Flexible libraries generated with 1-250 ng of Coriell NA12878 DNA were amplified with Normalase primers with the recommended number of PCR cycles to yield ≥ 12 nM, confirmed by qPCR. Libraries were either normalized with Normalase or (B) manually diluted to 4 nM based on the qPCR quantification and pooled. **C:** both pools were sequenced on individual Illumina MiSeq v2 Standard flowcells 1x50 cycles. **B:** The Normalase normalized libraries loaded at 12 pM clustered at 892 K/mm² and the index balance variation was calculated to be a CV of 5.6%. **C:** qPCR manually normalized libraries loaded at 10 pM clustered at 848 K/mm² and the index balance variation was calculated to be a CV of 23%. Both library pools performed within specifications of MiSeq clustering but Normalase provided 4-fold better library balancing.



Specifications

Specification	Feature
Post-Library amplification molarity required prior to Normalase	≥ 12 nM in 20 µl library volume
Normalized library concentration post Normalase	4 nM
Library balance within a pool	Coefficient of variation ≤ 10%
Library compatibility	<ul style="list-style-type: none">Libraries with full-length indexed adapters (e.g. Swift 2S, 2S Turbo Flexible, and kits from KAPA® and Illumina)Libraries that have an amplified yield of consistently ≥ 12 nM in 20 µL volumeLibraries prepared for direct sequencing (i.e., whole genome, whole transcriptome)Target enriched library pools post-hybridization capture that have indexed adapters
Swift Indexing kit compatibility	<ul style="list-style-type: none">2S with Set A, Set B, Set S1-S4, Set A MID, Set B MID, and Set S1-S4 MID single or dual indexed adapters2S Turbo Flexible with indexed adapters from Illumina or IDT
System compatibility	Reproducible performance across Illumina sequencing instruments
Kit size	96 reactions

Ordering Information

Product Name	Reactions	Catalog No.
Swift Normalase Kit	96	66096

Visit www.swiftbiosci.com for easy ordering.



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