

TailorMix Ready-to-Use Dual-Indexed PhiX Control Library, Denatured Catalog # TM-502

Introduction

SeqMatic's dual-indexed PhiX control library is used as a control for Illumina sequencing instruments. Our custom designed dual-indexed library eliminates contamination of PhiX sequencing reads in sample data, which enables the generation of cleaner raw data in multiplexed sequencing run.

Features

- **Designed for MiSeq, MiniSeq, NextSeq and HiSeq 2500:** Can be used in place of Illumina PhiX V3 in all Illumina systems.
- **High diversity:** Contains randomized inserts with 45% GC content and free of adapter dimers*. Can be used to supplement low diversity sequencing runs.
- **Unique dual index barcode:** Custom designed index sequences compatible with all Illumina sample prep kits.
- **Easy to use:** Supplied as a ready-to-use 20pM single stranded library in HT1 buffer for immediate spike in.

Ordering Information

Catalog #	Supplied Volume
TM-502	0.2 mL

Note: Denatured PhiX is not compatible with platforms using ExAmp cluster generation (HiSeq X and HiSeq 3000/4000).

Non-denatured PhiX is available for ExAmp cluster generation platforms TailorMix Dual-Indexed PhiX Control Library, non-denatured

- Compatible for all Illumina Sequencing platforms, including HiSeq3000/4000, HiSeq X.
- 10nM non-denatured library supplied in TE, pH8.0.

Catalog #	Supplied Volume
TM-580	10 μ L

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*No detectable adapter dimer in sequencing data bioinformatically.

Contents

- Sample type: Single stranded Illumina adapter ligated library
- Buffer condition: Illumina HT1 buffer
- Concentration: 20 pM
- Insert size: 380 bp

Storage Recommendations

The TailorMix Dual-Indexed PhiX Control Library is stable when stored at -20°C. Storage at -80°C is recommended for long term storage.

Best Practices

- Do not store the TailorMix Ready-to-Use Dual-Indexed PhiX Control Library in frost-free freezers.
- Aliquots should be made to allow for use to minimize repeated freeze/thaw cycles.
- Determine the appropriate aliquot volume based on usage.
- Aliquots that are in use can be stable for up to one month if kept on ice at all times during use and stored at temperature of -20°C or lower when not in use.

Consumables Preparation

Please make sure all consumables and equipment are available before starting this experiment.

Consumables and Equipment	Supplier
Micropipettor	General lab supplier
Micropipettor Tips	General lab supplier
HT1 Buffer	Illumina
Microcentrifuge tubes	General lab supplier

Procedures

1. Denature and dilute your sequencing libraries according to Illumina standard operating procedures.
2. Heat up the TailorMix Ready-to-Use Dual-Indexed PhiX Control Library at 94°C for 3 min before combining it with the denatured and diluted sequencing library.
3. Combine the 20pM TailorMix Ready-to-Use Dual-Indexed PhiX Control Library with your denatured and diluted sequencing library.
 - a. **For standard TruSeq and Nextera libraries:**
 - i. Add 1% of the denatured TailorMix Ready-to-Use Dual-Indexed PhiX Control Library to your sample as a positive control for QC purposes.
 - b. **For miRNA and Small RNA libraries:**
 - i. Add 1% of the denatured TailorMix Ready-to-Use Dual-Indexed PhiX Control Library to your sample as a positive control for QC purposes.
 - c. **For low diversity amplicon libraries:**
 - i. Add 15-25% of the denatured TailorMix Ready-to-Use Dual-Indexed PhiX Control Library to your sample as a positive control for QC purposes and for increasing diversity of your samples.

Note: *Optimal ratio of TailorMix Ready-to-Use Dual-Indexed PhiX Control Library in the sequencing run could be different for each amplicon library.*

 - d. **For dedicated control lanes on HiSeq:**
 - i. Load the proper concentration of denatured TailorMix Ready-to-Use Dual-Indexed PhiX Control Library according to Illumina's standard operating procedures for your instrument.
4. Load your sample onto the sequencing or clustering instrument according to Illumina's standard operating procedures for your instrument.

Sequencing Sample Sheet Setup

The TailorMix Dual-Indexed PhiX control library contains dual-indexed barcodes which can be demultiplexed with all Illumina compatible demultiplexing software. To monitor the percentage of PhiX reads in a sequencing run, include the TailorMix Dual-Indexed PhiX barcodes on the sequencing Sample Sheet for demultiplexing.

If the TailorMix Dual-Indexed PhiX barcodes are not included on the sequencing Sample Sheet, the PhiX reads will be parsed into a folder named “Unknown” or “Undetermined” along with any other reads with index sequences not defined in your sample sheet.

For individual downstream analysis of the Dual-Indexed PhiX library, your sample sheet can be modified to parse the PhiX reads into its own individual set of files. Simply add a new line to your sample sheet file containing the sequences below as your index barcodes.

For single index read sequencing runs, only use the i7 Index 1 barcode sequence. The sequence length may also be adjusted as necessary to match the length of the index read.

	i7 Index 1 barcode	i5 Index 2 barcode	
	<ul style="list-style-type: none"> All Illumina systems 	<ul style="list-style-type: none"> MiSeq HiSeq 2000/2500 	<ul style="list-style-type: none"> MiniSeq NextSeq
8 bp	TCGAATGA	GACATGCG	CGCATGTC
6 bp	TCGAAT	GACATG	CGCATG

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