

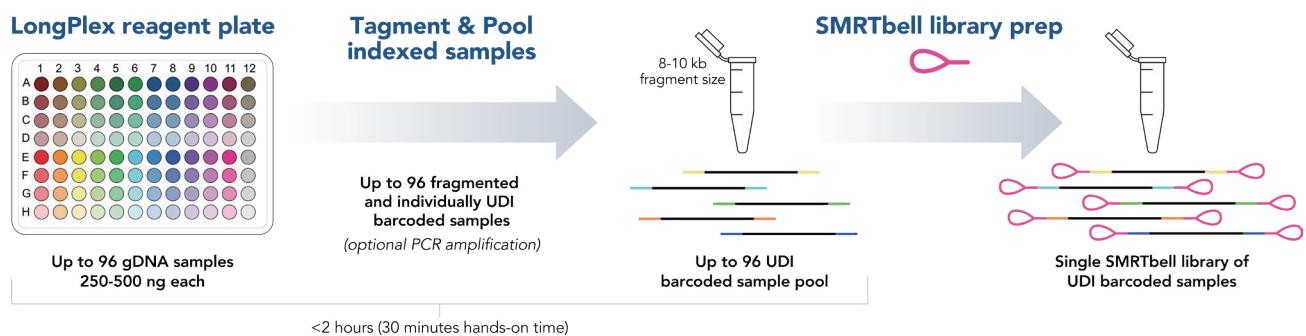
LongPlex™ Long Fragment Multiplexing Kit

Realize the throughput and cost-effectiveness of PacBio™ HiFi sequencing using seqWell's LongPlex Long Fragment Multiplexing Kit. Its speed, simplicity, and scalability enable massive sample multiplexing that unleashes scalable long read sequencing.

Supported Long-Read Applications:

- Microbial and small genome sequencing
- Metagenomics
- Low pass sequencing
- Targeted hybrid capture

LongPlex Workflow Upstream of SMRTbell Library Prep

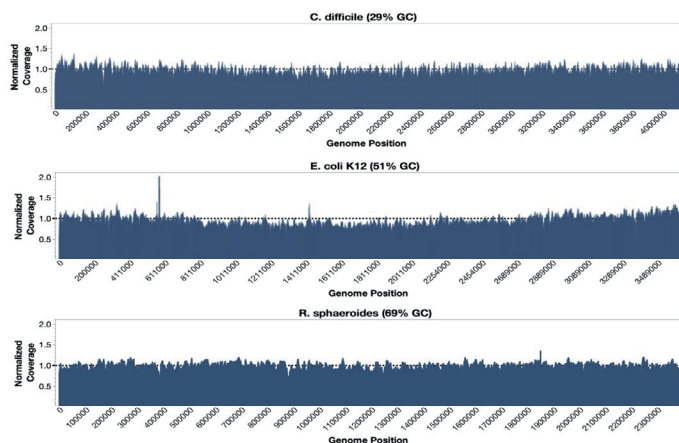


Key Benefits: Realize the potential of your PacBio long read sequencer

- **SPEED:** Eliminate mechanical shearing using a fully scalable, enzymatic method to simultaneously fragment & tag genomic DNA resulting in typical fragment sizes ranging from 8 – 10 kb
- **SIMPLICITY:** Automation-friendly, plate-based method with total workflow performed in <2 hours (30 minutes hands-on time) upstream of SMRTbell library preparation
- **SCALABILITY:** Massive multiplexing using 96 unique dual indexes (UDI) that can be combinatorially expanded with PacBio SMRTbell indexing
- **SAVINGS:** Early sample pooling greatly reduces all-in cost per sample for long read sequencing without sacrificing data quality
- **FLEXIBILITY:** PCR-free and PCR-based protocols available to support a variety of applications and sample needs

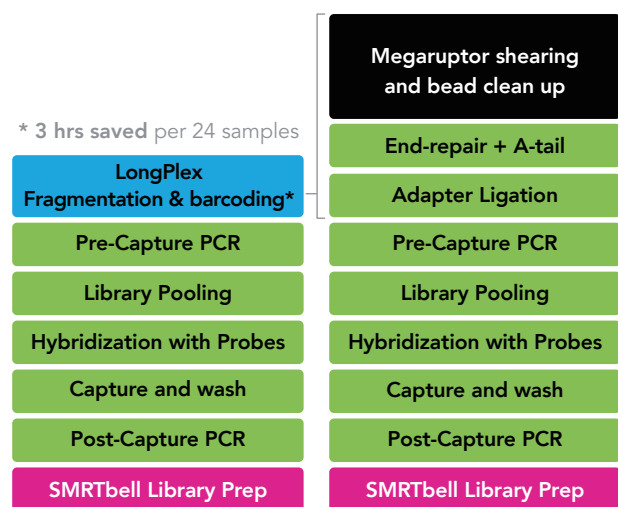
Application: Microbial Whole Genome Sequencing

- Consistent insert size across different genomes and different DNA input levels
- Uniform coverage across genomes with high, medium and low GC-contents
- Equal volume pooling upstream of PacBio SMRTbell library preparation - no quantification or normalization required to obtain even read counts



Organism	%GC	Genome Size (Mb)	PCR-Free Coverage	PCR-Free Max Contig (Mb)
<i>Clostridioides difficile</i>	29	4.3	160	4.1
<i>Staphylococcus epidermis</i>	32	2.6	396	2.6
<i>Bacillus cereus</i>	35	5.4	74	5.4
<i>Bacillus subtilis</i>	44	4.2	23	1.2
<i>Escherichia coli</i>	51	4.6	191	4.6
<i>Enterobacter cloacae</i>	55	5.3	136	5.3
<i>Bordetella pertussis</i>	67	4.0	259	4.0
<i>Rhodobacter sphaeroides</i>	69	4.5	209	3.2

Microbial gDNA samples from 8 different microbial isolate strains (ATCC), which contained GC contents ranging from 29-69%, were processed in quadruplicate using the LongPlex Long Fragment Multiplexing Kit. Post LongPlex tagging, samples were pooled into 4 x 24-plexes for bead-based size selection followed by PacBio SMRTbell prep kit 3.0. The 4 SMRTbell libraries were pooled into a single Revio SMRT Cell.



Application: Targeted Hybrid Capture

- Streamlined, automation-friendly workflow
- Save up to 3 hours/24 samples compared to Megaruptor fragmentation
- Equivalent data to mechanically sheared DNA
- Compatible with a variety of hybrid capture panels including Twist PGx and Dark Gene panels

Plex Name	Reads per sample	HiFi read length (bp)	% Duplication	Mean Target Coverage	Fold80 Penalty
8-plex capture A	207,095	4505	18.0%	177.6	1.76
8-plex capture B	159,129	4515	13.4%	147.6	1.79
8-plex capture C	118,512	4790	9.6%	109.0	1.84
8-plex capture D	162,724	4749	12.8%	154.6	1.81

32 samples were prepared with LongPlex using varying inputs (250-500 ng) of NA12878. Samples were pooled into 4 x 8-plexes post LongPlex, and then captured using a Twist PGx panel following manufacturer's protocol. Post capture samples underwent SMRTbell library preparation and were sequenced on the PacBio Revio.



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