

Application		Whole Genome Sequencing					RNA Sequencing		Metagenomics		Targeted Sequencing			
		De Novo Assembly - HiFi Reads	De Novo Assembly - Long Reads	De Novo Assembly - for Low DNA Input	Microbial De Novo Assembly	Variant Detection	Structural Variation Detection	Iso-Seq Method	Single-Cell Iso-Seq	Full-length 16S rRNA Sequencing	Shotgun Metagenomic Profiling or Assembly	Amplicon Sequencing	No-Amp Targeted Sequencing	
<b>Experimental Design</b>														
Experimental Design	<b>With 1 SMRT Cell 8M you can:</b>	Produce reference quality assemblies for genomes up to 2 Gb	Produce reference quality assemblies for genomes up to 3 Gb	Produce reference quality assemblies for genomes up to 1 Gb Multiplex up to 2 small genomes on the Sequel II System	Sequence up to 48 microbes	With 2 SMRT Cells 8M, Call SNVs, InDels, and SVs in a 3 Gb genome	Call SVs for up to 2 samples with ~3 Gb genomes	Characterize alternative splicing/annotate a genome with full length transcripts	Characterize alternative splicing with full length transcripts up to 3M full length reads with cell barcode and UMI information	Multiplex up to 96 samples to provide strain level resolution	Generate near-complete assemblies of high-complexity sample(s) (e.g. gut microbiome)	Sequence 384 barcoded amplicons	Sequence 5 targeted regions in a multiplex of 10 samples	
	<b>Minimum Recommended Coverage</b>	>15-fold HiFi read coverage	≥30-fold Unique Molecular Coverage (UMC) per haplotype	≥30-fold UMC per haplotype	≥30-fold UMC coverage per microbial genome	>15-fold HiFi of a human genome	<a href="#">5- to 25-fold Unique Molecular Coverage (UMC) coverage depending on study goals</a>	One human transcriptome per SMRT Cell 8M	1,000 unique reads/ single cell for 3000 cells 10,000 unique reads/ single cell for 300 cells	8,000 reads/sample	See Best practices guide	30-fold ≥Q20 CCS read coverage for variant detection 6,000-fold ≥Q20 CCS read coverage for minor variant detection (1% sensitivity)	≥100-fold ≥Q20 CCS read coverage per target locus	
	<b>Library Insert Size</b>	15 - 20 kb	>30 kb	~20 kb	10 - 15 kb	15 - 20 kb	>15 kb	<2 kb to >3 kb	<2 kb to >3 kb	1 - 2 kb	10 kb	500 bp - 15 kb	4-6 kb or larger	
<b>Sample Preparation</b>														
SMRTbell Template Preparation	<b>Procedure and Checklist Reference</b>	<a href="#">Preparing HiFi SMRTbell Libraries using SMRTbell Express Template Prep Kit 2.0</a>	<a href="#">Preparing gDNA Libraries Using the SMRTbell Express Template Preparation Kit 2.0</a>	<a href="#">Preparing SMRTbell Libraries Using Express Template Prep Kit 2.0 With Low DNA Input</a>	<a href="#">Preparing Multiplexed Microbial Libraries Using SMRTbell Express Template Prep Kit 2.0</a>	<a href="#">Preparing HiFi SMRTbell Libraries using SMRTbell Express Template Prep Kit 2.0</a>	<a href="#">Preparing gDNA Libraries Using the SMRTbell Express Template Preparation Kit 2.0</a>	<a href="#">Iso-Seq Express Template Preparation for Sequel and Sequel II Systems</a>	<a href="#">Preparing Single-Cell Iso-Seq Libraries Using SMRTbell Express Template Prep Kit 2.0</a>	<a href="#">Amplification of Full-Length 16S Gene with Barcoded Primers for Multiplexed SMRTbell Library Preparation and Sequencing</a>	<a href="#">Preparing 10 kb Library Using SMRTbell Express Template Prep Kit 2.0 for Metagenomics Shotgun Sequencing</a>	<a href="#">Preparing SMRTbell Libraries using PacBio Barcoded Overhang Adapters for Multiplex SMRT Sequencing</a>	<a href="#">No-Amp-Targeted-Sequencing-Utilizing-the-CRISPR-Cas9-System</a>	
	<b>Minimum Input Amount</b>	15 µg	≥1 µg for 10 kb ≥3 µg for >15 kb ≥5 µg for >30 kb	150 ng per 300 Mb genome size	1 µg per microbe	15 µg	3 µg	300 ng total RNA for 1st Strand cDNA Synthesis	>160 ng cDNA AFTER reamplification	500 ng - 1 µg	1.5 µg	250-500 ng for 250-1000 bp 500-1000ng for 1-3 kb bp 1000-2000 ng for 3-10 kb 3000 ng for 15kb	5 to 10 µg (represented by either a single sample or the total of multiple samples that will be multiplexed)	
	<b>Recommended PacBio Template Prep Kit</b>	Express TPK 2.0 + SMRTbell Enzyme Cleanup kit + Sequencing primer v2	Express TPK 2.0 + Sequencing primer v4	Express TPK 2.0 + Sequencing primer v4	Express TPK 2.0 + Sequencing primer v4	Express TPK 2.0 + SMRTbell Enzyme Cleanup kit + Sequencing primer v2	Express TPK 2.0 + Sequencing primer v2	Express TPK 2.0 + Sequencing primer v4	Express TPK 2.0 + Sequencing primer v4	Express TPK 2.0 + Sequencing primer v4	Express TPK 2.0 + Sequencing primer v2	Express TPK 2.0 + Sequencing primer v4	No-Amp Accessory Kit	
	<b>Multiplexing/SMRT Cell</b>	N/A	N/A	N/A	Up to 48 microbes / SMRT Cell 8M Up to 16 microbes / SMRT Cell 1M	N/A	Up to 2 human samples/ SMRT Cell 8M N/A SMRT Cell 1M	The protocol supports up to 12 barcodes available.	Detects cell barcodes and UMIs	Up to 96 samples/ SMRT Cell 8M Up to 12 samples/ SMRT Cell 1M	Profile up to 4 communities/ SMRT Cell 8M Profile one community/ SMRT Cell 1M	Up to 1,000+ samples/ SMRT Cell 8M or SMRT Cell 1M	Up to 10 samples/SMRT Cell	
<b>SMRT Sequencing with the Sequel II System: Loading and Pre-Extension Recommendations</b>														
Sequencing Preparation and Yield Performance	<b>Sequel II Binding Kit</b>	2.0	2.0	2.0	2.0	2.0	2.0	2.0 / 2.1*	2.0 / 2.1*	2.1	2.0	2.1: 500bp - 3,000 bp 2.0: ≥3,000 bp	2.0	
	<b>Sequel II Sequencing Plate</b>	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	
	<b>Sequencing Mode</b>	CCS	CLR	CLR / CCS	CLR	CCS	CLR	CCS	CCS	CCS	CCS	CCS	CCS	CCS
	<b>Movie Collection Time</b>	30 h	15 h	15 / 30 h	15 h	30 h	15 h	24 h	24 h	10 h	30 h	Insert Size-Dependent	≥10 h (20 h for repeat expansion targets)	
	<b>Notes</b>	*Use Binding Kit 2.1 as default and 2.0 for long transcripts												
<b>SMRT Sequencing with the Sequel System: Loading and Pre-Extension Recommendations</b>														
Sequencing Preparation and Yield Performance	<b>Sequel Binding kit</b>	3.0	3.0	3.0	3.0	3.0	3.0	3.0	3.0	3.0	3.0	3.0	3.0	
	<b>Sequel Sequencing Plate</b>	3.0	3.0	3.0	3.0	3.0	3.0	3.0	3.0	3.0	3.0	3.0	3.0	
	<b>Sequencing Mode</b>	CCS	CLR	CLR	CLR	CCS	CLR	CCS	CCS	CCS	CCS	CCS	CCS	
	<b>Movie Collection Time</b>	20 h	10 h	10 h	10 h	20 h	10 h	20 h	20 h	10 h	30 h	Insert Size-Dependent	≥10 h (20 h for repeat expansion targets)	
	<b>Notes</b>													
<b>Data Analy</b>														
Other Compatible SoTools (Contact FAS for)	<b>SMRT Analysis GUI Applications</b>	CCS	Assembly (HGAP 4)	Assembly (HGAP 4)	Demultiplex Barcodes followed by Microbial Assembly analysis	CCS with Mapping	Structural Variant Calling	Iso-Seq	Iso-Seq	CCS	CCS	CCS or Long Amplicon Analysis	Demultiplex Barcodes followed by CCS with Mapping analysis	
	<a href="#">PacBio Devnet Tools</a>	Falcon, Falcon Unzip, Falcon Phase	Falcon, Falcon Unzip, Falcon Phase										Repeat Analysis Tools	
	<b>Notes</b>	Recommend: GATK, Google Deep Variant <a href="#">Github Tools</a>												

Read lengths, reads/data per SMRT Cell and other sequencing performance results vary based on sample quality/type and insert size

[Pac Bio Glossary of Terms](#)

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