

PARAMETER	BASIC SPECS OF SEQUENCING PLATFORMS <sup>1</sup>				
	NovaSeq6000	HiSeq3000	NextSeq500	MiSeq	PacBioSEQUEL
Chemistry	SBS reverse terminators	SBS reverse terminators	SBS reverse terminators	SBS reverse terminators	Direct SBS, Single molecule seq, cleavable fluor (gammaP)
Seq Configurations	Flow cells: S4 (4 lanes) and S2, S1 and SP (2 lanes)	Eight independent loading ports per flow cell	Single loading port per flow cell run	Single loading port per flow cell run	8 independent SMRT cells per run
Most common error	Substitutions, G-C bias	Substitutions, G-C bias	Substitutions, G-C bias	Substitutions, G-C bias	Insertions GC deletions
% Error rate <sup>3</sup>	~0.1	~0.1	~0.1	~0.1	CLR (12-15), CCS (~0.1)
Seq Read length	35-150 bp 250 bp (SP only)	35-150 bp	35-150 bp	35-300 bp	Large-insert: 15-25 kb Amplicon, IsoSeq: 30-45 kb
SE Reads/full run	Full S4 flow cell: 10.0 G Full S2 flow cell: 4.1 G Full S1 flow cell: 1.6 G Full SP flow cell: 0.8 G	Full flow cell: 2.4 G One lane: 300 M	400 M	10-13 M (version 2 chem) 20-25 M (version 3 chem)	450-600k
Max output/ PE run	6000 Gb	720 Gb	120 GB	~15 Gb	Large-insert genomic: 15 Gb Amplicon, IsoSeq: 30 Gb
Run time	1-4 days	1-3 days	1-2 days	1-3 days	10h- 20h/ cell
<sup>4</sup> Cost/Gb (\$)	8 to 23 (S4 to SP) <sup>4</sup>	25.1	42.5	127.4	57.3
Most common app	Whole genome Variant Analysis Transcriptome (RNAseq) Digital GE ChIP seq, SeqCap, ExomeSeq Methylation Anal (WGBS, RRBS)	Whole genome Variant Analysis Transcriptome (RNAseq) Digital GE ChIP seq, SeqCap, ExomeSeq Methylation Anal (WGBS, RRBS)	Whole genome Variant Analysis Transcriptome (RNAseq) Digital GE ChIP seq, SeqCap, ExomeSeq Methylation Anal (WGBS, RRBS)	Amplicon (e.g., 16S metagenomics) Targeted RNAseq Variant Analysis	<i>De novo</i> sequencing Isoform analysis (IsoSeq) Amplicon Seq SNP validation Direct methyl-seq Microbial seq Variant analysis

<sup>1</sup>Table prepared by David Moraga at the Univ. of Florida-ICBR Core facilities, June 2019. These data may be different from the specs provided by instrument vendors. Many of the numbers reflect our own experience.

<sup>2</sup>Fragment coverage increased with A-T content.

<sup>3</sup>Percentage error within a single read of the maximum length.

<sup>4</sup>Based on average price of highest output run configuration at the University of Florida Core facility (e.g., one lane, 1/8 region, one SMRT cell). Prices at other facilities may be significantly different. Compare to Sanger Seq of 0.2-0.5 cents/base.

**Abbreviations:** SBS= sequencing by synthesis, SE= single end, PE= paired-end, CLR= continuous length read, CCS= circular consensus sequence (Hi-fi), WGBS= whole-genome bisulfite sequencing, RRBS= reduced-representation bisulfite sequencing, SeqCap= sequence capture, ChIP= chromatin immunoprecipitation, GE= Gene Expression.