

The future of simple benchtop NGS is here

Sequencing Archer™ NGS solutions on the Illumina MiSeq™ i100 Series

The Archer portfolio of next generation sequencing (NGS) solutions from Integrated DNA Technologies was designed to be tech-friendly and simple, just like the MiSeq i100 Series from Illumina. By pairing the two, labs can now generate high-quality targeted RNA- and DNA-sequencing data fast for cancer and hereditary disease research.

To demonstrate sequencing performance for Archer VARIANTPlex™ and FUSIONPlex™ NGS Solutions on the Illumina MiSeq i100 Series, a cross-site study was designed to assess sequencer run metrics and resulting variant data from 13 panels across 12 input materials.

The results of this study demonstrate repeatable, consistent sequencer run metrics and highly correlative variant calling data for fusions, single nucleotide variants (SNVs) and insertions and deletions (Indels) across sites and platforms (Illumina MiSeq i100 Series versus Illumina NextSeq 500/550 Systems).

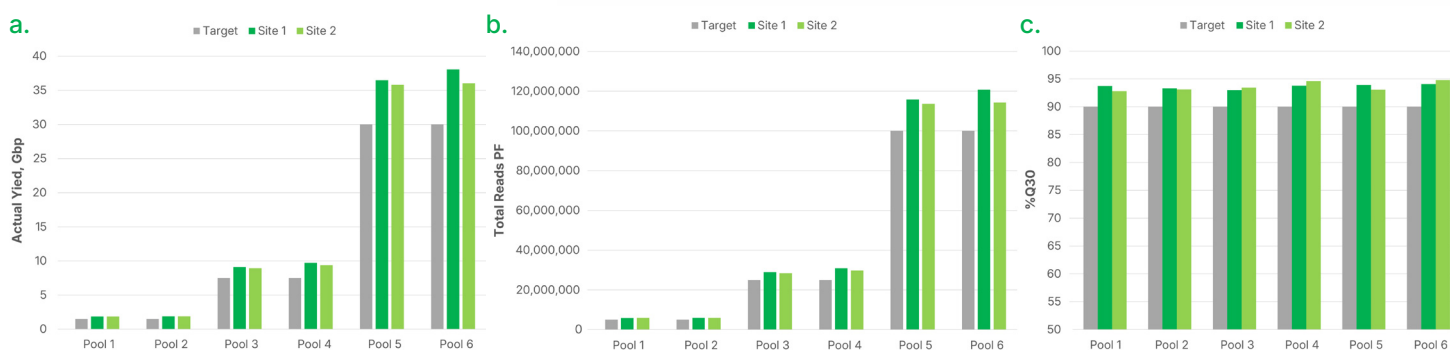
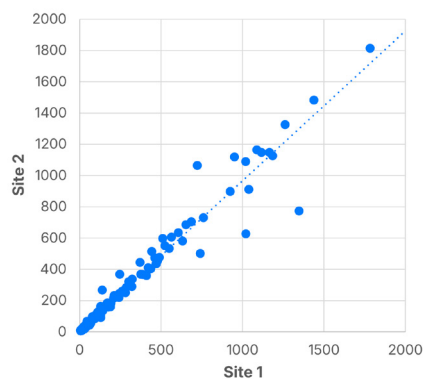


Figure 1: Following manufacturer recommendations for sample library pool preparation (RA-DOC-054 Rev 05, RA-DOC-620 Rev 02) and sequencer loading (#200055785 v02) Actual Yield (a), Total Reads Passing Filter (b), and Q30 scores (c) universally exceeded target metrics across two geographically separate sites, two operators, and twelve sequencing runs. Target metrics in gray and passing metrics in green.

a. Unique fusion supporting reads *



b. Variant Allele Frequency

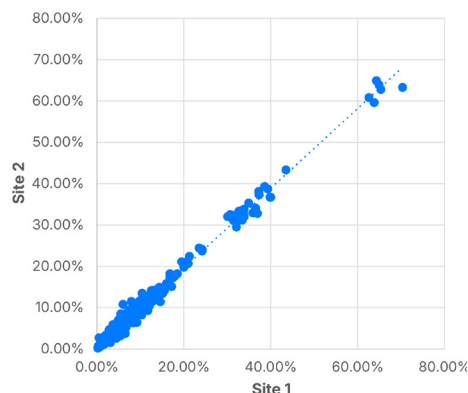
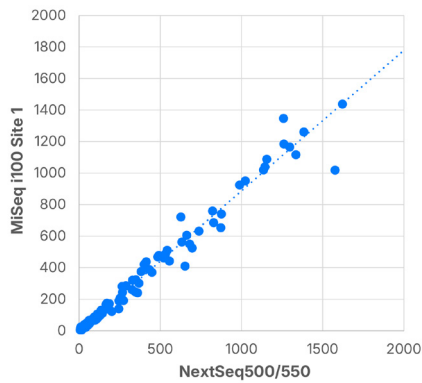


Figure 2: 100% of expected fusions (a) and SNV/Indels (b) were called at or above analytical limit of detection previously observed. Correlations between unique supporting reads ($R^2 = 0.97$) and variant allele frequency ($R^2 = 0.99$) are shown. The different marker colors represent unique inputs of various qualities ranging in mass from 10ng-200ng across 5 FUSIONPlex panels (a) and 8 VARIANTPlex panels (b).

* n = 1 fusion not shown at (2934, 2793) reads for image readability

a. Fusion Supporting Reads*



b. Variant Allele Frequency Correlation

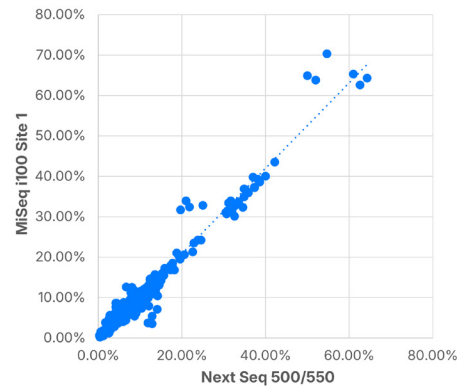


Figure 3: Fusion (a) and SNV/Indel (b) calls were highly correlated between MiSeq i100 Series and NextSeq 500/550 datasets generated from the same pools - unique supporting reads ($R^2 = 0.98$) and variant allele frequency ($R^2 = 0.96$) are shown.

* n= 1 fusion not shown at (2934, 3101) reads for image readability

Table 1: Detailed sequencer run metrics for Site 1 (a) and Site 2 (b)

a.

Pool	Kit	Q30	Clusters PF	Percent Occupied	Actual Yield, Gbp	Targeted PhiX	PhiX Actual	Total Reads PF	Quoted Reads After Demux
1	5M	93.72%	74.2%	82.23	1.85	2%	1.44%	5,870,000	109%
2	5M	93.28%	75.4%	83.2	1.88	2%	1.9%	5,962,103	112%
3	25M	92.98%	73.11%	81.85	9.11	2%	1.43%	28,904,965	103%
4	25M	93.78%	78.13%	88.3	9.74	2%	1.08%	30,891,919	116%
5	100M	93.91%	73.22%	83.16	36.48	2%	1.46%	115,798,935	108%
6	100M	94.05%	76.36%	86.71	38.06	2%	1.37%	120,766,891	114%

b.

Pool	Kit	Q30	Clusters PF	Percent Occupied	Actual Yield, Gbp	Targeted PhiX	PhiX Actual	Total Reads PF	Quoted Reads After Demux
1	5M	92.80%	75.04%	85.1	1.87	2%	1.74%	5,934,006	110%
2	5M	93.12%	75.20%	82.4	1.88	2%	2.54%	5,946,614	111%
3	25M	93.42%	71.67%	79.6	8.93	2%	2.60%	28,337,103	100%
4	25M	94.61%	73.34%	84.1	9.38	2%	2.24%	29,785,456	111%
5	100M	93.07%	71.87%	83.0	35.83	2%	2.25%	113,656,483	105%
6	100M	94.76%	72.30%	80.3	36.04	2%	3.10%	114,339,842	106%



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