



NGI-Stockholm -- Science For Life Laboratory Best-practice analysis for quality checking report A.Blomberg_15_01 -- P1887_101

For sample P1887_101 belonging to the project A.Blomberg_15_01 NGI-Stockholm best-practice analysis for quality checking has been performed. For mate pair libraries produced with Nextera, best-practice analysis described at this address has been performed:

http://res.illumina.com/documents/products/technotes/technote_nextera_matepair_data_processing.pdf

The following tools have been employed (tools are listed in order of execution):

- j trimmomatic:/proj/a2010002/nobackup/sw/mf/bioinfo-tools/misc/trimmomatic/0.30/trimmomatic-0.30.jar
- ij fastqc:/sw/apps/bioinfo/fastqc/0.10.1/milou/fastqc
- jij abyss:/sw/apps/bioinfo/abyss/1.3.5/milou/bin/

The results from each tool is reported in the following sections. Moreover you will find all the results and commands that have been run in the delivery folder on Uppmax

Trimmomatic

Reads (both paired and mate pairs) can contain parts of the adapter sequence or, in the case of mate pairs, part of the linker sequence. Illumina recommends to remove the adapter before use of the reads in any downstream analysis (this is mandatory for mate pairs).

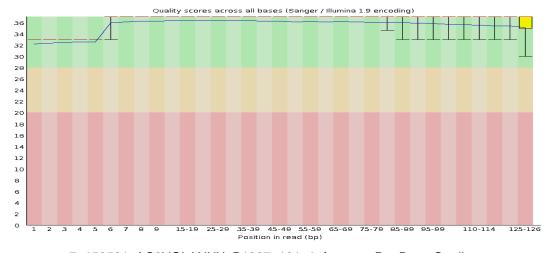
Adapter sequences removed are:

- TACACTCTTTCCCTACACGACGCTCTTCCGATCT
- ii GTGACTGGAGTTCAGACGTGTGCTCTTCCGATCT
- iii AATGATACGGCGACCACCGAGATCTACACTCTTTCCCTACACGACGCTCTTCCGATCT
- iv CAAGCAGAAGACGCATACGAGATCGGTCTCGGCATTCCTGCTGAACCGCTCTTCCGATCT
- V AATGATACGGCGACCACCGAGATCTACACTCTTTCCCTACACGACGCTCTTCCGATCT

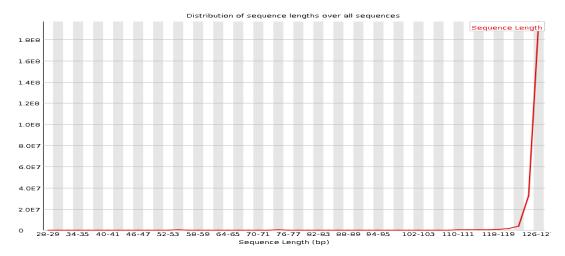
P1887_101	#orig_pairs	#survived_pairs
7_150521_AC6VGLANXX_P1887_101	265653467	246861333 (93%)
total	265653467	246861333 (93%)

P1887_101	#survived_fw_only	#survived_rv_only	#discarded
7_150521_AC6VGLANXX_P1887_101	16895870 (6%)	1358485 (1%)	537779 (0%)
total	16895870 (6%)	1358485 (1%)	537779 (0%)

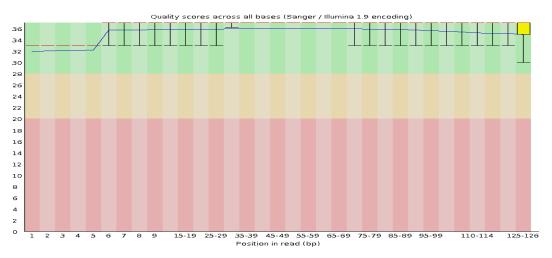
Fastqc



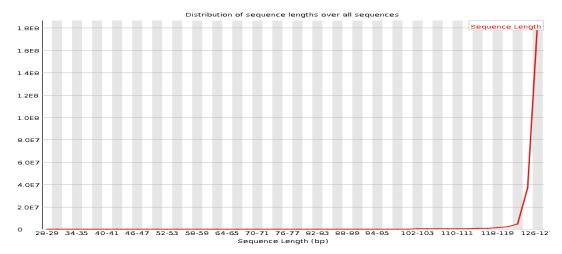
7_150521_AC6VGLANXX_P1887_101_1_fastqc -- Per Base Quality



7_150521_AC6VGLANXX_P1887_101_1_fastqc -- Sequence Length Distribution



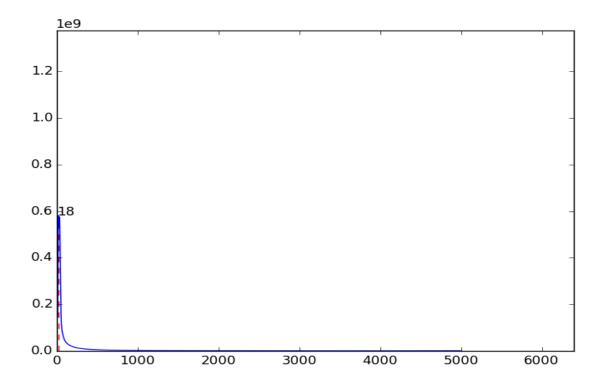
7_150521_AC6VGLANXX_P1887_101_2_fastqc -- Per Base Quality



7_150521_AC6VGLANXX_P1887_101_2_fastqc -- Sequence Length Distribution

Abyss

A possible way to assess the complexity of a library even in absence of a reference sequence is to look at the kmer profile of the reads. The idea is to count all the kmers (i.e., sequence of length k) that occur in the reads. In this way it is possible to know how many kmers occur 1,2,..., N times and represent this as a plot. This plot tell us for each x, how many k-mers (y-axis) are present in the dataset in exactly x-copies. In an ideal world (no errors in sequencing, no bias, no repeating regions) this plot should be as close as possible to a gaussian distribution. In reality we will always see a peak for x=1 (i.e., the errors) and another peak close to the expected coverage. If the genome is highly heterozygous a second peak at half of the coverage can be expected.



kmer profile with k=35.