# Manual for trim\_fastq2.pl

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## Summary:

The Perl program trim\_fastq.pl takes fastq files of single or paired-end sequence reads and trims lowquality bases from the 5' and 3' ends, also it removes any reads that contain at least one "N" (undefined base) after trimming. Trimming is done from the 5' and 3' ends until the first base with a quality score above a certain threshold is reached or the read gets shorter than a certain minimum length. In case of paired-end reads, pairs where both reads meet the criteria are kept in a pairs file, and in case only one read of a pair meets the criteria, this read is kept in a separate singlets file.

Usage: trim\_fastq2.pl <options> <file(s)>

# Options: *-a* type of analysis:

- s for single reads
- t for paired-end reads with pairs in two files
- *p* for paired-end reads with pairs in one file
- -o offset for the quality scores (default: 33, for certain types of Illumina pipelines it can be 64, then this option must be set)
- -/ minimum length of read that is kept (default: 40)
- -q minimum base quality that must be reached at the 3' endfor the trimming to stop (default: 10)
- -r core name for results file(s). For paired-ends, will be extended by \_pairs.fastq, for single-ends by \_singlets.fastq, default trim\_fastq. Must contain only letters, numbers, or \_

Important: Bases and quality sequences must be only ONE line each (i.e. four lines containing the information for each read).

### **Output files:**

#### trim\_fastq\_ singlets.fastq:

when trimming single reads: all reads that were kept

when trimming paired-end reads: all reads that meet the quality criteria but where the mate does not meet the quality criteria

#### trim\_fastq\_ pairs.fastq

only written when trimming paired-end reads, contains all read pairs that meet the quality criteria, in format *pair1\_read1 pair1\_read2 pair2\_read1 pair2\_read2* etc.

## trim\_fastq\_output\_stats.txt

output statistics, gives an overview over the number of input reads, the number of reads that were kept in the single and paired files and the number of reads of length x that were kept

#### **Examples:**

trim single reads in file reads.fastq, output reads\_trimmed\_singlets.fastq and reads\_trimmed\_output\_stats.txt

perl trim fastq.pl -a s reads.fastq -r reads trimmed

trim single reads in file reads.fastq with offset 64, minimum quality score at 3' end 20, and minimum length of 50

perl trim\_fastq.pl -a s -o 64 -l 50 -q 20 reads.fastq

trim paired-end reads with both reads in file read\_pairs.fastq (reads that belong to one pair must be directly after each other, the file must contain an even number of reads, i.e. no mixture of single and paired-end reads)

perl trim fastq.pl -a p reads pairs.fastq

trim paired-end reads with the first read of each pair in file read1.fastq and the second read in each pair in file read2.fastq (reads must be in the same order in the files)

perl trim\_fastq.pl -a t reads1.fastq read2.fastq

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