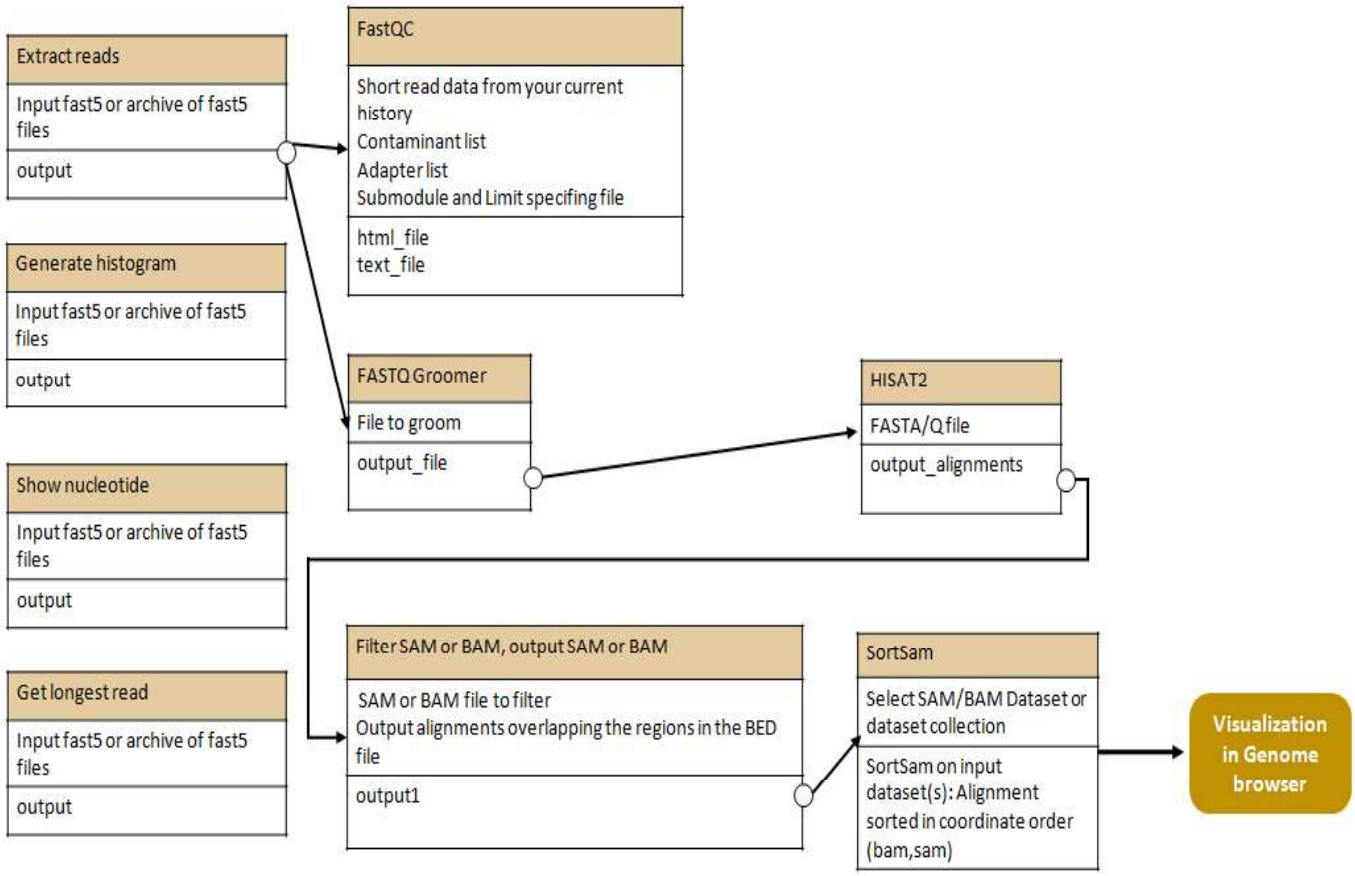

Figure 1. NGS pipeline for Cas13 mediated transcript targeting and cleavage. The complete pipeline was built on Galaxy v 19.01 an open, web-based platform for biological data analysis. Briefly, .fast5 sequence files generated from Nanopore sequencing (MiOxford Nanopore Technologies (ONT), Oxford, UK.) were uploaded in the Galaxy platform. The sequence files were extracted (Extract Reads) for read-lengths (Generate histogram), read-distribution (Show nucleotide) and getting the longest read from the set of .fast5 files (Get longest read). The .fast5 files were parallelly analyzed for the quality of the reads (FastQC) and converted into .fastqsanger format suitable for the downstream mapping programs (FASTQ Groomer). The .fastqsanger files thus generated were mapped onto the Human genome (GRCh38.p12, GRCh38/hg38) by HISAT2 alignment program compatible for RNA-seq. The mapped files generated as .bam and .sam files were further filtered with Map quality Score (MAPQ) (Filter SAM or BAM, output SAM or BAM) to eliminate unmapped reads. The filtered alignment files were further sorted in the order of coordinates (SortSam). The alignments were finally visualized in Integrative Genomics Viewer version 2.5. 2. Finally, cleavage points in the cells treated gRNA-lshCas13 were determined in reference to the cells that lacked plasmid insert.