

## **- Abstract and Introduction:**

- Long-read sequencing offers a number of advantages over short-read sequencing.
- Although Short-reads are effective in cost, accurate, and supported by analysis tools and pipelines, sequencing nucleic acid polymers in short fragments is difficult.
- Long-read technologies are good at accuracy and generate reads in excess of 10 kb, so the characteristics of long-read data should be focused on the analysis tools.
- This paper will study error correction, base modification detection, and long-read transcriptomics analysis and the challenges.
- It will Describe the principles of long-read data analysis.
- It also introduces open-source catalogue of long-read analysis tools: [long-read-tools.org](http://long-read-tools.org).
- short-read sequencers such as Illumina's NovaSeq, HiSeq, NextSeq, and MiSeq instruments ; BGI's MGISEQ and BGISEQ models ; or Thermo Fisher's Ion Torrent sequencers.
- long-read sequencing technologies such as Pacific Biosciences' (PacBio) single-molecule real-time (SMRT) sequencing and Oxford Nanopore Technologies' (ONT) nanopore sequencing.
- Long-reads can also improve de novo assembly, mapping certainty, transcript isoform identification, and detection of structural variants.
- we check available tools to deal with long-read sequencing projects.

## **- Related Work:**

- I) Pollard MO, Gurdasani D, Mentzer AJ, Porter T, Sandhu MS. Long reads: their purpose and place. *Hum Mol Genet.* 2018; 27(R2):234–41. <https://doi.org/10.1093/hmg/ddy177>.
- II) Burgess DJ. Genomics: next regeneration sequencing for reference genomes. *Nat Rev Genet.* 2018; 19(3):125. <https://doi.org/10.1038/nrg.2018.5>.
- III) Bentley DR, Balasubramanian S, Swerdlow HP, Smith GP, Milton J, Brown CG, et al. Accurate whole human genome sequencing using reversible terminator chemistry. *Nature.* 2008; 456(7218):53–9. <https://doi.org/10.1038/nature07517>.
- IV) Goodwin S, McPherson JD, McCombie WR. Coming of age: ten years of next-generation sequencing technologies. *Nat Rev Genet.* 2016; 17(6):333–51. <https://doi.org/10.1038/nrg.2016.49>.
- V) Jeon SA, Park JL, Kim J-H, Kim JH, Kim YS, Kim JC, et al. Comparison of the MGISEQ-2000 and Illumina HiSeq 4000 sequencing platforms for RNA sequencing. *Genomics Inform.* 2019; 17(3):e32.
- VI) Rothberg JM, Hinz W, Rearick TM, Schultz J, Mileski W, Davey M, et al. An integrated semiconductor device enabling non-optical genome sequencing. *Nature.* 2011; 475(7356):348–52. <https://doi.org/10.1038/nature10242>.
- VII) Quail M, Smith ME, Coupland P, Otto TD, Harris SR, Connor TR, et al. A tale of three next generation sequencing platforms: comparison of Ion torrent, Pacific Biosciences and Illumina MiSeq sequencers. *BMC Genomics.* 2012; 13(1):341. <https://doi.org/10.1186/1471-2164-13-341>.

- There are open-source static catalogues (e.g. [github.com/B-UMMI/long-read-catalog](https://github.com/B-UMMI/long-read-catalog)), custom pipelines developed by individual labs for specific purposes, and others that attempt to generalise them for a wider research community.
- For this purpose, we introduce [long-read-tools.org](https://long-read-tools.org), a timely database that comprehensively collates tools used for long-read data analysis.
- In addition to true long-read sequencing technologies (SMRT and nanopore), we include synthetic long-read strategies. We include them in our database for completeness but indicate when they have been superseded or are no longer maintained.
- [long-read-tools.org](https://long-read-tools.org) is an open-source project under the MIT License, whose code is available through GitHub [161].

## **– Result:**

- Nanopore reads-based transcriptomes are more recent , and work is still needed to understand the characteristics of these data (e.g. coverage bias, sequence biases, reproducibility). Certain isoform assembly pipelines predict a large number of unannotated isoforms requiring validation and classification.
- Long reads theoretically confer huge advantages over short reads for transcript-level differential expression, however the low-level of replication and modest read counts obtained from long-read transcriptomic experiments are currently limiting.
- Until throughput increases and price decreases sufficiently, hybrid approaches that use long reads to define the isoforms expressed in the samples and short reads to get enough counts for well-powered differential expression may be successful; these do not yet exist.