**Comparison of circRNA tools for detection of circular RNA in total RNA-Seq**

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| **Algorithm** | **Language** | **Mapper** | **Requirements** | **Filter** | **Strength** | **Weakness** |
| **Indirect and Multi-Stage Approaches (Segmented read-based approach)** | | | | | | |
| MapSplice  (2010) | Python | Bowtie | **-** Upmapped sequencing reads  - Paired/Single-end | GTF-based alignment, score-based | - High accuracy  - Good sensitivity | - Slow  - Requires annotated genome  - High disk space usage |
| find\_circ  (2013) | Python | Bowtie2 | - Expects not-mapping reads  - Single-end | Manual output filtering, count-based | - Fast  - Low RAM requirements | - Low accuracy |
| CIRCfinder  (2013) | Python | TopHat2,  Bowtie | - Limited to intronic circRNAs | Filtering by supplied intronic regions | - Designed for ciRNAs | - Does not work for exonic circRNAs |
| CircExplorer  (2014) | Pyhton | TopHat2,  Bowtie, (STAR) | - Unmapped sequencing reads  - Paired/Single-end | Manual output filtering, count-based | - High accuracy  - Good sensitivity  - Good balance between precision and sensitivity  - Requires low memory | - Slow  - Requires annotated genome |
| UROBORUS  (2016) | Perl | TopHat2, Bowtie | - Expects not-mapping reads | Manual output filtering, count-based | - Optimized for total rRNA-data  - Low RAM requirements | - Low Sensitivity  - no *de novo* circRNAs |
| **Approached Directly Employing Chimeric Reads** | | | | | | |
| circRNA\_finder  (2014) | Perl | STAR | - Unmapped sequencing reads  - Paired/Single-end | Minimum circRNA length | - Fast | - Low sensitivity  - Low accuracy |
| Segemehl (2014) | C | segemehl  (internal aligner) | - Unmapped sequencing reads  - Paired/Single-end | Alignment accuracy, multiple hits | - Fast  - High Sensitivity | - High RAM requirements for indexing  - Low precision |
| CIRI  (2015) | Perl | BWA | - Reads premapped with BWA  - Paired/Single-end | Stringency filter exon colinearity | - High sensitivity  - Good balance between precision and sensitivity | - Slow  - High RAM requirements  - Low accuracy |
| DCC  (2016) | Python | STAR | - Unmapped sequencing reads  - Paired/Single-end | GTF-based, count-cased, gene-based | - Improved detection of small circRNA  - Integrated into circtools workflow | - High RAM usage for large genomes |
| CircExplorer2  (2016) | Python | Multiple | - Unmapped sequencing reads | Manual output filtering, count-based | - Multiple read mappers supported | - Mandatory genome annotation |
| **Tools Using Statistical Approaches (candidate-based approaches)** | | | | | | |
| KNIFE  (2015) | Python, Perl, R | Bowtie, Bowtie2 | - Unmapped sequencing reads  - Paired/Single-end | Posterior probability or p-value | - Good balance between precision and sensitivity  - Explits read and mapping quality | - Mandatory genome annotation |
| CIRI2  (2018) | Perl | BWA | - Reads pre-mapped with BWA | Stringency filter, exon colinearity | - High sensitivity | - High RAM requirements  - Slow  - Low accuracy |
| Acfs  (2016) | Perl | BWA | - Expects not-mapping reads | circRNA length, splice-strength | - Detects non-canonical splicing | - Paired-end data perfomrance |
| NCLScan |  |  |  |  | - High Precision  - Low RAM requirements |  |
| PTESFinder |  |  |  |  |  | - High RAM requirements |

**Comparison of circRNA workflows**

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| --- | --- | --- | --- | --- | --- |
| **Workflow** | **Description + Features** | **Characteristics** | **Requirements** | **Strength** | **Weakness** |
| circtools (2018) | * one-stop software solution for circular RNA research * circRNA detection, internal sequence reconstruction, quality checking, statistical testing, screening for enrichment of RBP binding sites, differential exon RNase R resistance, circRNA primer design | * **Detection module**: based on DCC * **Reconstruct module:** read mapping using STAR and detection of back splice junctions * **Output:** read counts for detected circRNA, coordinated linearCount and CircSkipJunctions | * **Input:** Unmapped sequencing reads (fastq files) * **Language:** Python3 and Python2 * **External tools:**   + STAR (for detection and reconstruction)   + Bedtools (for the enrichment)   + R (for visualization and primer design) | * allows circRNA prediction and an automated analysis for further characterization * fully integrated software suite * *de novo* detection of new exons * several different analyses * well documented | * needs unmapped sequencing reads (does not work with BAM files) * developed for Linux |
| CIRI  (2016) | * CircRNA detection, annotation, *de novo* assembly of circRNA transcripts, alternative splicing analyses | * **Detection module**: based on CIRI | * **Input:** Sequencing reads mapped with BWA * **Language:** Perl * **External tools:**   + BWA   + Perl | * easy to install, few dependencies | * no validation module * only BWA data supported |
| CircExplorer2 (2016) | * CircRNA detection, annotation, *de novo* assembly of circRNA transcripts, alternative splicing analyses | * **Detection module**: based on CircExplorer | * **Input:** Unmapped sequencing reads * **Language:** Python * **External tools:**   + STAR   + Bedtools | * fully integrated software suite * well documented | * several external dependencies * no validation modules |

# References

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