

Package ‘Rfastp’

March 26, 2024

Type Package

Title An Ultra-Fast and All-in-One Fastq Preprocessor (Quality Control, Adapter, low quality and polyX trimming) and UMI Sequence Parsing).

Version 1.12.0

Description Rfastp is an R wrapper of fastp developed in c++. fastp performs quality control for fastq files. including low quality bases trimming, polyX trimming, adapter auto-detection and trimming, paired-end reads merging, UMI sequence/id handling. Rfastp can concatenate multiple files into one file (like shell command cat) and accept multiple files as input.

License GPL-3 + file LICENSE

Encoding UTF-8

LazyData true

RoxygenNote 7.1.1

biocViews QualityControl, Sequencing, Preprocessing, Software

SystemRequirements GNU make

LinkingTo Rcpp, Rhtslib, zlibbioc

Imports Rcpp, rjson, ggplot2, reshape2

Suggests BiocStyle, testthat, knitr, rmarkdown

VignetteBuilder knitr

git_url <https://git.bioconductor.org/packages/Rfastp>

git_branch RELEASE_3_18

git_last_commit 16a614b

git_last_commit_date 2023-10-24

Repository Bioconductor 3.18

Date/Publication 2024-03-25

Author Wei Wang [aut] (<<https://orcid.org/0000-0002-3216-7118>>),
Ji-Dung Luo [ctb] (<<https://orcid.org/0000-0003-0150-1440>>),
Thomas Carroll [cre, aut] (<<https://orcid.org/0000-0002-0073-1714>>)

Maintainer Thomas Carroll <tc.infomatics@gmail.com>

R topics documented:

| | |
|-------------|----|
| catfastq | 2 |
| curvePlot | 3 |
| qcSummary | 4 |
| rfastp | 5 |
| trimSummary | 11 |

| | |
|--------------|-----------|
| Index | 12 |
|--------------|-----------|

| | |
|----------|---------------------------------|
| catfastq | <i>Concatenate Fastq Files.</i> |
|----------|---------------------------------|

Description

concatenate multiple fastq files into a single file.

Usage

```
catfastq(output, inputFiles, append = FALSE, paired = FALSE, shuffled = FALSE)
```

Arguments

| | |
|------------|--|
| output | output file name [string] |
| inputFiles | a vector of input file names [vector] |
| append | a logical indicating append the files to a file already exists. |
| paired | a logical indicating split the input files into two halves. the first half merged into read1, the second half merged into read2. |
| shuffled | a logical indicating split the input file list into two halves. The R1/R2 files are interleaved in the inputFiles vector. |

Value

no returns.

Author(s)

Wei Wang

Examples

```
pe001_read1 <- system.file("extdata", "splited_001_R1.fastq.gz",
  package="Rfastp")
pe002_read1 <- system.file("extdata", "splited_002_R1.fastq.gz",
  package="Rfastp")
pe003_read1 <- system.file("extdata", "splited_003_R1.fastq.gz",
  package="Rfastp")
pe004_read1 <- system.file("extdata", "splited_004_R1.fastq.gz",
```

```
    package="Rfastp")

pe001_read2 <- system.file("extdata", "splited_001_R2.fastq.gz",
  package="Rfastp")
pe002_read2 <- system.file("extdata", "splited_002_R2.fastq.gz",
  package="Rfastp")
pe003_read2 <- system.file("extdata", "splited_003_R2.fastq.gz",
  package="Rfastp")
pe004_read2 <- system.file("extdata", "splited_004_R2.fastq.gz",
  package="Rfastp")

allR1 <- c(pe001_read1, pe002_read1, pe003_read1, pe004_read1)
allR2 <- c(pe001_read2, pe002_read2, pe003_read2, pe004_read2)

allreads <- c(allR1, allR2)
allreads_shuffled <- c(pe001_read1, pe001_read2, pe002_read1, pe002_read2,
  pe003_read1, pe003_read2, pe004_read1, pe004_read2)

outputPrefix <- tempfile(tmpdir = tmpdir())
# a normal concatenation for single-end libraries.

catfastq(output = paste0(outputPrefix, "_R1.fastq.gz"), inputFiles = allR1)

# a normal concatenation for paired-end libraries.

catfastq(output = paste0(outputPrefix, "merged_paired"),
  inputFiles = allreads, paired=TRUE)

# Append to exist files (paired-end)

catfastq(output=paste0(outputPrefix,"append_paired"), inputFiles=allreads,
  append=TRUE, paired=TRUE)

# Input paired-end files are shuffled.

catfastq(output=paste0(outputPrefix,"_shuffled_paired"),
  inputFiles=allreads_shuffled, paired=TRUE, shuffled=TRUE)
```

curvePlot

Plot of Base Quality and GC Content.

Description

generate a ggplot2 object of Base Quality/GC content before and after QC.

Usage

```
curvePlot(json, curves = "quality_curves")
```

Arguments

json the output json of function rfastq. [json]
curves plots for Base Quality("quality_curves") or GC content("content_curves"). default is "quality_curves"

Value

a ggplot2 object.

Author(s)

Wei Wang

Examples

```
outputPrefix <- tempfile(tmpdir = tmpdir())  
se_read1 <- system.file("extdata", "Fox3_Std_small.fq.gz", package="Rfastp")  
se_json_report <- rfastp(read1 = se_read1, outputFastq = outputPrefix,  
  thread = 4)  
# Base Quality plot is the default output:  
p1 <- curvePlot(se_json_report)  
p1  
  
p2 <- curvePlot(se_json_report, curves = "content_curves")
```

qcSummary

Summary of Fastq Quality Control

Description

generate a data frame of the Fastq QC summary.

Usage

```
qcSummary(json)
```

Arguments

json the output json of function rfastq. [json]

Value

a data frame.

Author(s)

Wei Wang

Examples

```
outputPrefix <- tempfile(tmpdir = tmpdir())
se_read1 <- system.file("extdata", "Fox3_Std_small.fq.gz", package="Rfastp")
se_json_report <- rfastp(read1 = se_read1, outputFastq = outputPrefix,
  thread = 4)
df_summary <- qcSummary(se_json_report)
```

rfastp

R wrap of fastp

Description

Quality control (Cut adapter, low quality trimming, polyX trimming, UMI handling, and etc.) of fastq files.

Usage

```
rfastp(
  read1,
  read2 = "",
  outputFastq,
  unpaired = "",
  failedOut = "",
  merge = FALSE,
  mergeOut = "",
  phred64 = FALSE,
  interleaved = FALSE,
  fixMGId = FALSE,
  adapterTrimming = TRUE,
  adapterSequenceRead1 = "auto",
  adapterSequenceRead2 = "auto",
  adapterFasta = "",
  trimFrontRead1 = 0,
  trimTailRead1 = 0,
  trimFrontRead2 = 0,
  trimTailRead2 = 0,
  maxLengthRead1 = 0,
  maxLengthRead2 = 0,
  forceTrimPolyG = FALSE,
  disableTrimPolyG = FALSE,
  minLengthPolyG = 10,
  trimPolyX = FALSE,
  minLengthPolyX = 10,
  cutWindowSize = 4,
  cutLowQualTail = FALSE,
  cutSlideWindowRight = FALSE,
```

```

cutLowQualFront = FALSE,
cutMeanQual = 20,
cutFrontWindowSize = 4,
cutFrontMeanQual = 20,
cutTailWindowSize = 4,
cutTailMeanQual = 20,
cutSlideWindowSize = 4,
cutSlideWindowQual = 20,
qualityFiltering = TRUE,
qualityFilterPhred = 15,
qualityFilterPercent = 40,
maxNfilter = 5,
averageQualFilter = 0,
lengthFiltering = TRUE,
minReadLength = 15,
maxReadLength = 0,
lowComplexityFiltering = FALSE,
minComplexity = 30,
index1Filter = "",
index2Filter = "",
maxIndexMismatch = 0,
correctionOverlap = FALSE,
minOverlapLength = 30,
maxOverlapMismatch = 5,
maxOverlapMismatchPercentage = 20,
umi = FALSE,
umiLoc = "",
umiLength = 0,
umiPrefix = "",
umiSkipBaseLength = 0,
umiNoConnection = FALSE,
umiIgnoreSeqNameSpace = FALSE,
overrepresentationAnalysis = FALSE,
overrepresentationSampling = 20,
splitOutput = 0,
splitByLines = 0,
thread = 2,
verbose = TRUE
)

```

Arguments

| | |
|-------------|--|
| read1 | read1 input file name(s). [vector] |
| read2 | read2 input file name(s). [vector] |
| outputFastq | string of /path/prefix for output fastq [string] |
| unpaired | for PE input, output file name for reads which the mate reads failed to pass the QC [string], default NULL, discard it. [string] |
| failedOut | file to store reads that cannot pass the filters default NULL, discard it. [string] |

| | |
|----------------------|--|
| merge | for PE input, A logical(1) indicating whether merge each pair of reads into a single read if they are overlapped, unmerged reads will be write to 'output' file. Default is FALSE. the 'mergeOut' must be set if TRUE. |
| mergeOut | under 'merge' mode, file to store the merged reads. [string] |
| phred64 | A logical indicating whether the input is using phred64 scoring (it will be converted to phred33, so the output will still be . phred33) |
| interleaved | A logical indicating whether <read1> is an interleaved FASTQ which contains both read1 and read2. Default is FALSE. |
| fixMGIid | the MGI FASTQ ID format is not compatible with many BAM operation tools, enable this option to fix it. Default is FALSE |
| adapterTrimming | A logical indicating whether run adapter trimming. Default is 'TRUE' |
| adapterSequenceRead1 | the adapter for read1. For SE data, if not specified, the adapter will be auto-detected. For PE data, this is used if R1/R2 are found not overlapped. |
| adapterSequenceRead2 | the adapter for read2 (PE data only). This is used if R1/R2 are found not overlapped. If not specified, it will be the same as <adapterSequenceRead1> |
| adapterFasta | specify a FASTA file to trim both read1 and read2 (if PE) by all the sequences in this FASTA file. |
| trimFrontRead1 | trimming how many bases in front for read1, default is 0. |
| trimTailRead1 | trimming how many bases in tail for read1, default is 0' |
| trimFrontRead2 | trimming how many bases in front for read2. If it's not specified, it will follow read1's settings |
| trimTailRead2 | trimming how many bases in tail for read2. If it's not specified, it will follow read1's settings |
| maxLengthRead1 | if read1 is longer than maxLengthRead1, then trim read1 at its tail to make it as long as maxLengthRead1 Default 0 means no limitation. |
| maxLengthRead2 | if read2 is longer than maxLengthRead2, then trim read2 at its tail to make it as long as maxLengthRead2. Default 0 means no limitation. If it's not specified, it will follow read1's settings. |
| forceTrimPolyG | A logical indicating force polyG tail trimming, trimming is only automatically enabled for Illumina NextSeq/NovaSeq . data. |
| disableTrimPolyG | A logical indicating disable polyG tail trimming. |
| minLengthPolyG | the minimum length to detect polyG in the read tail. 10 by default. |
| trimPolyX | A logical indicating force polyX tail trimming. |
| minLengthPolyX | the minimum length to detect polyX in the read tail. 10 by default. |
| cutWindowSize | the window size option shared by cutLowQualFront, cutLowQualTail, or cutSlideWindowRight. Range: 1~1000, default: 4 |
| cutLowQualTail | A logical indiccating move a sliding window from tail (3') to front, drop the bases in the window if its mean quality < threshold, stop otherwise. Default is 'FALSE' |

| | |
|----------------------|---|
| cutSlideWindowRight | A logical indicating move a sliding window from front to tail, if meet one window with mean quality < threshold, drop the bases in the window and the right part, and then stop. Default is 'FALSE' |
| cutLowQualFront | A logical indicating move a sliding window from front (5') to tail, drop the bases in the window if its mean quality < threshold, stop otherwise. Default is 'FALSE' |
| cutMeanQual | the mean quality requirement option shared by cutLowQualFront, cutLowQualTail or cutSlideWindowRight. Range: 1~36, default: 20 |
| cutFrontWindowSize | the window size option of cutLowQualFront, default to cutWindowSize if not specified. default: 4 |
| cutFrontMeanQual | the mean quality requirement option for cutLowQualFront, default to cutMeanQual if not specified. default: 20 |
| cutTailWindowSize | the window size option of cutLowQualTail, default to cutWindowSize if not specified. default: 4 |
| cutTailMeanQual | the mean quality requirement option for cutLowQualTail, default to cutMeanQual if not specified. default: 20 |
| cutSlideWindowSize | the window size option of cutSlideWindowRight, default to cutWindowSize if not specified. default: 4 |
| cutSlideWindowQual | the mean quality requirement option for cutSlideWindowRight, default to cutMeanQual if not specified. default: 20 |
| qualityFiltering | A logical indicating run quality filtering. Default is 'TRUE'. |
| qualityFilterPhred | the minimum quality value that a base is qualified. Default 15 means phred quality ≥ 15 is qualified. |
| qualityFilterPercent | Maximum percents of bases are allowed to be unqualified (0~100). Default 40 means 40% |
| maxNfilter | maximum number of N allowed in the sequence. read/pair is discarded if failed to pass this filter. Default is 5 |
| averageQualFilter | if one read's average quality score < 'averageQualFilter', then this read/pair is discarded. Default 0 means no requirement. |
| lengthFiltering | A logical indicating whether run length filtering. Default: TRUE |
| minReadLength | reads shorter than minReadLength will be discarded, default is 15. |
| maxReadLength | reads longer than maxReadLength will be discarded, default 0 means no limitation. |

| | |
|------------------------------|--|
| lowComplexityFiltering | A logical indicating whether to run low complexity filter. The complexity is defined as the percentage of base that is different from its next base ($\text{base}[i] \neq \text{base}[i+1]$). Default is 'FALSE' |
| minComplexity | the threshold for low complexity filter (0~100). Default is 30, which means 30% complexity is required. (int [=30]) |
| index1Filter | specify a file contains a list of barcodes of index1 to be filtered out, one barcode per line. |
| index2Filter | specify a file contains a list of barcodes of index2 to be filtered out, one barcode per line. |
| maxIndexMismatch | the allowed difference of index barcode for index filtering, default 0 means completely identical. |
| correctionOverlap | A logical indicating run base correction in overlapped regions (only for PE data), default is 'FALSE' |
| minOverlapLength | the minimum length to detect overlapped region of PE reads. This will affect overlap analysis based PE merge, adapter trimming and correction. 30 by default. |
| maxOverlapMismatch | the maximum number of mismatched bases to detect overlapped region of PE reads. This will affect overlap analysis based PE merge, adapter trimming and correction. 5 by default. |
| maxOverlapMismatchPercentage | the maximum percentage of mismatched bases to detect overlapped region of PE reads. This will affect overlap analysis based PE merge, adapter trimming and correction. Default 20 means 20% |
| umi | A logical indicating whether to preprocess unique molecular identifier (UMI). Default: 'FALSE' |
| umiLoc | specify the location of UMI, can be (index1/index2/read1/read2/per_index/per_read) |
| umiLength | length of UMI if the UMI is in read1/read2. |
| umiPrefix | an string indicating the following string is UMI (i.e. prefix=UMI, UMI=AATTCG, final=UMIAATTCG). Only letters, numbers, and '#' allowed. No prefix by default. |
| umiSkipBaseLength | if the UMI is in read1/read2, skip 'umiSkipBaseLength' bases following UMI, default is 0. |
| umiNoConnection | an logical indicating remove "_" between the UMI prefix string and the UMI string. Default is FALSE. |
| umiIgnoreSeqNameSpace | an logical indicating ignore the space in the sequence name. Default is FALSE, the umi tag will be inserted into the sequence name before the first SPACE. |
| overrepresentationAnalysis | A logical indicating overrepresentation analysis. Default is 'FALSE' |

| | |
|----------------------------|---|
| overrepresentationSampling | one in 'overrepresentationSampling' reads will be computed for overrepresentation analysis (1~10000), smaller is slower, default is 20. |
| splitOutput | number of files to be splitted (2~999). a sequential number prefix will be added to output name. Default is 0 (no split) |
| splitByLines | split output by limiting lines of each file(>=1000), a sequential number prefix will be added to output name (0001.out.fq, 0002.out.fq...), default is 0 (disabled). |
| thread | owrker thread number, default is 2 |
| verbose | output verbose log information |

Value

returns a json object of the report.

Author(s)

Thomas Carroll, Wei Wang

Examples

```
# prepare for the input and output files.
# if the output file exists, it will be OVERWRITEN.

se_read1 <- system.file("extdata", "Fox3_Std_small.fq.gz", package="Rfastp")
pe_read1 <- system.file("extdata", "reads1.fastq.gz", package="Rfastp")
pe_read2 <- system.file("extdata", "reads2.fastq.gz", package="Rfastp")
outputPrefix <- tempfile(tmpdir = tmpdir())

# a normal single-end file

se_json_report <- rfastp(read1 = se_read1,
  outputFastq=paste0(outputPrefix, "_se"), thread = 4)

# merge paired-end data by overlap:

pe_json_report <- rfastp(read1 = pe_read1, read2 = pe_read2, merge = TRUE,
  outputFastq = paste0(outputPrefix, '_unpaired'),
  mergeOut = paste0(outputPrefix, '_merged.fastq.gz'))

# a clipr example

clipr_json_report <- rfastp(read1 = se_read1,
  outputFastq = paste0(outputPrefix, '_clipr'),
  disableTrimPolyG = TRUE,
  cutLowQualFront = TRUE,
  cutFrontWindowSize = 29,
  cutFrontMeanQual = 20,
```

```
    cutLowQualTail = TRUE,  
    cutTailWindowSize = 1,  
    cutTailMeanQual = 5,  
    minReadLength = 29,  
    adapterSequenceRead1 = 'GTGTCAGTCACTTCCAGCGG'  
  )
```

trimSummary

Summary of Fastq adapter and low quality trimming

Description

generate a data frame of the Fastq trim summary.

Usage

```
trimSummary(json)
```

Arguments

json the output json of function rfastq. [json]

Value

a data frame.

Author(s)

Wei Wang

Examples

```
outputPrefix <- tempfile(tmpdir = tempdir())  
se_read1 <- system.file("extdata", "Fox3_Std_small.fq.gz", package="Rfastp")  
se_json_report <- rfastp(read1 = se_read1, outputFastq = outputPrefix,  
  thread = 4, adapterSequenceRead1 = 'GTGTCAGTCACTTCCAGCGG')  
trim_summary <- trimSummary(se_json_report)
```

Index

catfastq, [2](#)

curvePlot, [3](#)

qcSummary, [4](#)

rfastp, [5](#)

trimSummary, [11](#)