Package 'Motif2Site'

November 15, 2024

Type Package

Title Detect binding sites from motifs and ChIP-seq experiments, and compare binding sites across conditions

Version 1.10.0

Depends R (>= 4.1)

Description
Detect binding sites using motifs IUPAC sequence or bed coordinates and ChIP-seq experiments in bed or bam format. Combine/compare binding sites across experiments, tissues, or conditions. All normalization and differential steps are done using TMM-GLM method. Signal decomposition is done by setting motifs as the centers of the mixture of normal distribution curves.

BugReports https://github.com/ManchesterBioinference/Motif2Site/issues

License GPL-2

LazyData false

Encoding UTF-8

Imports S4Vectors, stats, utils, methods, grDevices, graphics, BiocGenerics, BSgenome, GenomeInfoDb, MASS, IRanges, GenomicRanges, Biostrings, GenomicAlignments, edgeR, mixtools

Suggests BiocStyle, rmarkdown, knitr, BSgenome.Hsapiens.UCSC.hg38, BSgenome.Mmusculus.UCSC.mm10, BSgenome.Scerevisiae.UCSC.sacCer3, BSgenome.Ecoli.NCBI.20080805

biocViews Software, Sequencing, ChIPSeq, DifferentialPeakCalling, Epigenetics, SequenceMatching

RoxygenNote 7.1.2

VignetteBuilder knitr

git_url https://git.bioconductor.org/packages/Motif2Site

git_branch RELEASE_3_20

git_last_commit 12ad0b4

git_last_commit_date 2024-10-29

Repository Bioconductor 3.20

Date/Publication 2024-11-14

Author Peyman Zarrineh [cre, aut] (<https://orcid.org/0000-0003-4820-4101>)

Maintainer Peyman Zarrineh <peyman.zarrineh@manchester.ac.uk>

Contents

`Bed2Granges` *Read a bed file as Genomic Ranges*

Description

Read a bed file as Genomic Ranges.

Usage

```
Bed2Granges(fileName)
```

Arguments

`fileName`	A table delimeted file in bed format

Value

granges format of given coordinates

Examples

```
yeastExampleFile=system.file("extdata", "YeastSampleMotif.bed",
     package="Motif2Site")
ex <- Bed2Granges(yeastExampleFile)
ex
```

`combine2Table` *Combine all IP and Input count table files*

Description

Open raw counts IP and Inut files and with given total counts calculate Fold Enrichment values, and combine them into one file

Usage

```
combine2Table(outputName, replicateNumber, currentDir)
```

Arguments

`outputName`	Name of the output table
`replicateNumber`	Number of the replicates
`currentDir`	Directory for I/O operations

Value

No return value

`combineMotifFiles` *Combine motif bed files into a combined ranges*

Description

Get motif file names and combine them into a matrix, and keep the indices of original motifs in the combined file.

If the motif type is string the bed files are deleted after being combined to one matrix.

Usage

```
combineMotifFiles(motifFileNames, motifType = "BioString")
```

Arguments

motifFileNames	a vector motif file names
motifType	Type of motif string or give bed

Value

No return value

`combineTestResults` *Combine count Table and statistics table*

Description

Combine count table and pvalue FE statistics into one file for motifs and regions seperately.

Usage

```
combineTestResults(
  motifFile,
  acceptedMotifsOutputFile,
  acceptedRegionsOutputFile,
  countTableFile,
  testTableFile,
  fdrCutoff,
  windowSize
)
```

Arguments

motifFile	File contains motifs
acceptedMotifsOutputFile	File name of accepted motif table inforation
acceptedRegionsOutputFile	File name of accepteted region information
countTableFile	Table of count values file name
testTableFile	negative binomial test table file name
fdrCutoff	Pvalue cut-off related to the used FDR
windowSize	Window size around binding site. The total region would be 2*windowSize+1

Value

The average binding intensity for each ChIP-seq

`compareBedFiless2UserProvidedRegions` *Compare a set of bed files to a user provided regions set*

Description

This function gets user provided bedfiles and compare them with a user provided region.

It returns this comparison to given user binding regions in terms of precision/recall.

Usage

```
compareBedFiless2UserProvidedRegions(bedfiles, motifnames, givenRegion)
```

Arguments

`bedfiles`	a vector of bed files
`motifnames`	a vector of the names related to bed files
`givenRegion`	granges of user provided binding regions

Value

A dataframe which includes precision recall values for each bed file

See Also

`compareMotifs2UserProvidedRegions`

Examples

```
yeastExampleFile=system.file("extdata", "YeastSampleMotif.bed",
                             package="Motif2Site")
YeastRegionsChIPseq <- Bed2Granges(yeastExampleFile)
bed1 <- system.file("extdata", "YeastBedFile1.bed", package="Motif2Site")
bed2 <- system.file("extdata", "YeastBedFile2.bed", package="Motif2Site")
BedFilesVector <- c(bed1, bed2)
SequenceComparison <- compareBedFiless2UserProvidedRegions(
     givenRegion=YeastRegionsChIPseq,
     bedfiles=BedFilesVector,
     motifnames=c("YeastBed1", "YeastBed2")
     )
SequenceComparison
```

`CompareBeds2GivenRegions` *Compare a set of bed files to a provided regions set*

Description

Get combined ranges of bed files and compare them to given

binding regions in terms of precision/recall.

Usage

```
CompareBeds2GivenRegions(motifName, bindingRegions)
```

Arguments

`motifName`	a vector of motif names
`bindingRegions`	granges of provided binding regions

Value

A dataframe which includes precision recall values for each motif

`CompareMotifs2GivenRegions` *Comparison motifs locations to a given regions set*

Description

Comparison of motifs locations to user provided binding regions.

It returns this comparison to given user binding regions in terms of precision/recall.

Usage

```
CompareMotifs2GivenRegions(motifs, mismatchNumbers, bindingRegions)
```

Arguments

`motifs`	a vector of motif characters in nucleotide IUPAC format
`mismatchNumbers`	a vector Number of mismatches allowed to match with motifs
`bindingRegions`	granges of user provided binding regions

Value

A dataframe which includes precision recall values for each motif

`compareMotifs2UserProvidedRegions` *Compare a set of motifs to a user provided regions set*

Description

This function gets user provided motifs and related mismatch numbers, it detects motifs and compare them with a user provided region.

It returns this comparison to given user binding regions in terms of precision/recall.

The genome and build information should be provided and relevant BS genomes packages such as BSgenome.Mmusculus.UCSC.mm10 or BSgenome.Hsapiens.UCSC.hg38 must be installed for the used genome and builds.

Usage

```
compareMotifs2UserProvidedRegions(
  motifs,
  mismatchNumbers,
  genome,
  genomeBuild,
  DB = "UCSC",
  givenRegion,
  mainCHRs = TRUE
)
```

Arguments

`motifs`	a vector of motif characters in nucleotide IUPAC format
`mismatchNumbers`	a vector Number of mismatches allowed to match with motifs
`genome`	The genome name such as "Hsapiens", "Mmusculus", "Dmelanogaster"
`genomeBuild`	The genome build such as "hg38", "hg19", "mm10", "dm3"
`DB`	The database of genome build. default: "UCSC"
`givenRegion`	granges of user provided binding regions
`mainCHRs`	If true only the major chromosome are considered, if FALSE Random, Uncharacterised, and Mithocondrial chromosomes are also considered

Value

A dataframe which includes precision recall values for each motif

See Also

`compareBedFiless2UserProvidedRegions`

Examples

```
# Artificial example in Yeast
# install BSgenome.Scerevisiae.UCSC.sacCer3 prior to run this code
 yeastExampleFile=system.file("extdata", "YeastSampleMotif.bed",
                              package="Motif2Site")
YeastRegionsChIPseq <- Bed2Granges(yeastExampleFile)
SequenceComparison <- compareMotifs2UserProvidedRegions(
   givenRegion=YeastRegionsChIPseq,
   motifs=c("TGATTSCAGGANT", "TGATTCCAGGANT", "TGATWSCAGGANT"),
   mismatchNumbers=c(1,0,2),
   genome="Scerevisiae",
   genomeBuild="sacCer3"
   )
SequenceComparison
```

`computeFoldEnrichment` *compute fold enrichment values for an experiment*

Description

Open raw counts IP and Inut files and with given total counts calculate Fold Enrichment values for the motifs

Usage

```
computeFoldEnrichment(
  ipCountFile,
  inputCountFile,
  ipTotalCount,
  inputTotalCount,
  outputName
)
```

Arguments

`ipCountFile`	File contains motifs count values for IP experiment
`inputCountFile`	File contains motifs count values for Input experiment
`ipTotalCount`	Total short reads number in IP experiment
`inputTotalCount`	Total short reads number in Input experiment
`outputName`	Name of the output table

Value

No return value

data *Synthetic datasets used in the package*

Description

Comparison Yeast synthetic motifs and binding sites: Two synthetic motif files in bed fromat are created to compare them with a synthetic binding site set in terms of precision and recall.

Fur binding sites detection in E. coli build NC_000913: Synthetic Fur ChIp-seq in E. coli was generated using real peaks published in Seo et al 2014. The ChIP-seq data are provided in bed format in fe and dpd condition and both contains two replicates. Synthetic Input ChIP-seq datasets were generated by randomly distributing short reads in E. coli genome. User provided candidate binding sites in bed format was generated by combining instances of "GWWTGANAA" motif with 1-mistmatch and "GWWTGAGAAT" with 2-mismatches in E. coli genome.

Format

Three bed files to compare user provided motifs and binding sites in Yeast. Seven bed files to compare Fur ChIP-seq binding sites in E.coli.

YeastBedFile1.bed The first synthetic motif set

YeastBedFile2.bed The second synthetic motif set

YeastSampleMotif.bed The synthetic binding region

FurMotifs.bed User provided Fur motif set in E. coli

FUR_fe1.bed Synthetic Fur ChIP-seq short reads in fe condition rep1

FUR_fe2.bed Synthetic Fur ChIP-seq short reads in fe condition rep2

FUR_dpd1.bed Synthetic Fur ChIP-seq short reads in dpd condition rep1

FUR_dpd2.bed Synthetic Fur ChIP-seq short reads in dpd condition rep2

Input1.bed The synthetic background Input ChIP-seq rep1

Input2.bed The synthetic background Input ChIP-seq rep2

Examples

```
## Data for examplex to compare user provided motifs and binding sites in Yeast

yeastExampleFile=system.file("extdata", "YeastSampleMotif.bed",
                             package="Motif2Site")
YeastRegionsChIPseq <- Bed2Granges(yeastExampleFile)
bed1 <- system.file("extdata", "YeastBedFile1.bed", package="Motif2Site")
bed2 <- system.file("extdata", "YeastBedFile2.bed", package="Motif2Site")


## Data for examples of binding site detection in E. coli

# FUR candidate motifs in NC_000913 E. coli
FurMotifs = system.file("extdata", "FurMotifs.bed", package="Motif2Site")

# ChIP-seq FUR fe datasets binding sites from user provided bed file
# ChIP-seq datasets in bed single end format

IPFe <- c(system.file("extdata", "FUR_fe1.bed", package="Motif2Site"),
```

```
          system.file("extdata", "FUR_fe2.bed", package="Motif2Site"))


# ChIP-seq FUR dpd datasets binding sites from user provided bed file
# ChIP-seq datasets in bed single end format

IPDpd <- c(system.file("extdata", "FUR_dpd1.bed", package="Motif2Site"),
           system.file("extdata", "FUR_dpd2.bed", package="Motif2Site"))


# ChIP-seq background

Inputs <- c(system.file("extdata", "Input1.bed", package="Motif2Site"),
            system.file("extdata", "Input2.bed", package="Motif2Site"))
```

`decomposeBindingSignal` *Decompose binding signal among accepted motifs*

Description

Gets motif locations and related short reads and select the motifs which are non-skewed: abs(skewness) < 0.3 and more short reads binds closer to site, and show strong binding after decomposition.

Decomposition is performed by using mixtools normalmixEM command fixing mu as motif locations.

Usage

```
decomposeBindingSignal(
  windowSize,
  replicateNumber,
  acceptedRegionsOutputFile = "BindingRegions",
  acceptedMotifsOutputFile = "BindingMotifsTable",
  currentDir
)
```

Arguments

`windowSize`	Window size around binding site. The total region would be 2*windowSize+1
`replicateNumber`	experiment replicate number
`acceptedRegionsOutputFile`	File name contains binding regions coordinates and related motifs
`acceptedMotifsOutputFile`	File name contains motifs coordinates and related information, Pvalue, FE, etc
`currentDir`	Directory for I/O operations

Value

motifStatistics Ratio of accepted motifs, rejected motifs due to skewnewss, and rejected motifs after decomposition

`DeleteMultipleFiles` *Delete a vector of files*

Description

Delete multiple give files as a vector of characters

Usage

```
DeleteMultipleFiles(files)
```

Arguments

`files`	a vector of files

Value

No return value

`deriveHeuristicBindingDistribution` *build heurisitc distribution around the binding sites*

Description

This function generates heuristic distribution of short reads around binding sites which do not need to deconvolve, total numer of short reads and window size as number of neucleotid around binding sites.

It fits a kernel to the distribution and return the distribution as output. The total sum of returned values is equal to one. It plots this kernel.

Also it calculates FRiPs (Fraction of Reads in Peaks) for each

ChIP-seq and returns it. FRiPs and kernel distributions are measures of goodness of ChIP-seq experiments and selected motifs.

Usage

```
deriveHeuristicBindingDistribution(
  chipSeq,
  averageBindings,
  windowSize,
  acceptedRegionsOutputFile = "BindingRegions",
  currentDir
)
```

Arguments

chipSeq	ChIP-seq aligned 1nt short reads
averageBindings	expected short reads number aligned to a random location of genes of given size
windowSize	Window size around binding site. The total region would be 2*windowSize+1
acceptedRegionsOutputFile	Accepted binding regions
currentDir	Directory for I/O operations

Value

FRiPs Fraction of Reads in Peaks

DetectBindingSites *Detect binding sites from motif*

Description

DETECT Binding sites with given motif and mismatch number as well genome/build, False Discovery Rate for a given experiment name.

This function is called by both DetectBindingSitesBed and DetectBindingSitesMotif with different input.

Usage

```
DetectBindingSites(
  From,
  BedFile,
  motif,
  mismatchNumber,
  chipSeq,
  genome,
  genomeBuild,
  DB = "UCSC",
  fdrValue = 0.05,
  windowSize = 100,
  GivenRegion = NA,
  currentDir
)
```

Arguments

From	Type of motif dataset either "Motif" or "Bed"
BedFile	Motif locations in bed format file
motif	motif characters in nucleotide IUPAC format
mismatchNumber	Number of mismatches allowed to match with motifs
chipSeq	ChIP-seq alignment both IP and background in 1nt bed format files
genome	The genome name such as "Hsapiens", "Mmusculus", "Dmelanogaster"

genomeBuild	The genome build such as "hg38", "hg19", "mm10", "dm3"
DB	The database of genome build. default: "UCSC"
fdrValue	FDR value cut-off
windowSize	Window size around binding site. The total region would be 2*windowSize+1
GivenRegion	granges of user provided binding regions
currentDir	Directory for I/O operations

Value

A list of FRiPs, sequence statistics, and Motif statistics

`DetectBindingSitesBed` *Detect binding sites from bed motif input*

Description

Takes user provied bed regions, and check for validity of them. Read bam or bed alignment files and convert to 1 nt bed and call detect binding site from 1nt bed.

Usage

```
DetectBindingSitesBed(
  BedFile,
  IPfiles,
  BackgroundFiles,
  genome,
  genomeBuild,
  DB = "UCSC",
  fdrValue = 0.05,
  expName = "Motif_Centric_Peaks",
  windowSize = 100,
  format = ""
)
```

Arguments

BedFile	Motif locations in bed format file
IPfiles	IP ChIP-seq alignment files
BackgroundFiles	Background ChIP-seq alignment files. Can be Input experimetn, DNA whole exctract, etc.
genome	The genome name such as "Hsapiens", "Mmusculus", "Dmelanogaster"
genomeBuild	The genome build such as "hg38", "hg19", "mm10", "dm3"
DB	The database of genome build. default: "UCSC"
fdrValue	FDR value cut-off
expName	The name of the output table
windowSize	Window size around binding site. The total region would be 2*windowSize+1
format	alignment format and should be one of these: "BAMPE", "BAMSE", "BEDPE", "BEDSE"

Value

peakCallingStatistics A list FRiPs, sequence statistics, and Motif statistics

See Also

DetectBindingSitesMotif

Examples

```
# FUR candidate motifs in NC_000913 E. coli
FurMotifs=system.file("extdata", "FurMotifs.bed", package="Motif2Site")

# ChIP-seq datasets in bed single end format
IPFe <- c(system.file("extdata", "FUR_fe1.bed", package="Motif2Site"),
        system.file("extdata", "FUR_fe2.bed", package="Motif2Site"))
Inputs <- c(system.file("extdata", "Input1.bed", package="Motif2Site"),
            system.file("extdata", "Input2.bed", package="Motif2Site"))
FURfeBedInputStats <-
  DetectBindingSitesBed(BedFile=FurMotifs,
                        IPfiles=IPFe,
                        BackgroundFiles=Inputs,
                        genome="Ecoli",
                        genomeBuild="20080805",
                        DB="NCBI",
                        expName="FUR_Fe_BedInput",
                        format="BEDSE"
                       )
```

DetectBindingSitesMotif

Detect binding sites from sequence motif sequence and mismatchNumber

---

Description

DETECT Binding sites with given motif and mismatch number as well genome/build, False Discovery Rate for a given experiment name. Read bam or bed alignment files and convert to 1 nt bed and detect binding site among motifs from 1nt bed alignment.

Usage

```
DetectBindingSitesMotif(
  motif,
  mismatchNumber,
  IPfiles,
  BackgroundFiles,
  genome,
  genomeBuild,
  DB = "UCSC",
  fdrValue = 0.05,
```

```
  expName = "Motif_Centric_Peaks",
  windowSize = 100,
  format = "",
  GivenRegion = NA
)
```

Arguments

motif	motif characters in nucleotide IUPAC format
mismatchNumber	Number of mismatches allowed to match with motifs
IPfiles	IP ChIP-seq alignment files
BackgroundFiles	Background ChIP-seq alignment files. Can be Input experimetn, DNA whole exctract, etc.
genome	The genome name such as "Hsapiens", "Mmusculus", "Dmelanogaster"
genomeBuild	The genome build such as "hg38", "hg19", "mm10", "dm3"
DB	The database of genome build. default: "UCSC"
fdrValue	FDR value cut-off
expName	The name of the output table
windowSize	Window size around binding site. The total region would be 2*windowSize+1
format	alignment format and should be one of these: "BAMPE", "BAMSE", "BEDPE", "BEDSE"
GivenRegion	granges of user provided binding regions

Value

A list FRiPs, sequence statistics, and Motif statistics

See Also

DetectBindingSitesBed

Examples

```
# ChIP-seq datasets in bed single end format
IPFe <- c(system.file("extdata", "FUR_fe1.bed", package="Motif2Site"),
        system.file("extdata", "FUR_fe2.bed", package="Motif2Site"))
Inputs <- c(system.file("extdata", "Input1.bed", package="Motif2Site"),
            system.file("extdata", "Input2.bed", package="Motif2Site"))

 # Granages region for motif search
   NC_000913_Coordiante <-
     GenomicRanges::GRanges(seqnames=S4Vectors::Rle("NC_000913"),
                            ranges=IRanges::IRanges(1, 4639675))

FURfeStringInputStats <-
  DetectBindingSitesMotif(motif="GWWTGAGAA",
   mismatchNumber=1,
   IPfiles=IPFe,
   BackgroundFiles=Inputs,
   genome="Ecoli",
   genomeBuild="20080805",
```

```
  DB="NCBI",
  expName="FUR_Fe_StringInput",
  format="BEDSE",
  GivenRegion=NC_000913_Coordiante
  )
```

`DetectFdrCutoffBH` *FDR cut-off detection Benjamini Hochberg method*

Description

Return FDR cut-off for a user provided fdrvalue using Benjamini Hochberg on main motif test data

Usage

```
DetectFdrCutoffBH(TestTableFile = "TestResults", fdrValue = 0.05)
```

Arguments

`TestTableFile`	test table which contains pvalues
`fdrValue`	FDR cut-off

Value

pvalue cut-off

`findMotifs` *Find motif instances with a certain mismatch number*

Description

Find motif instances in a given genome. It gets motif strings and related allowed mismatchnumbers and returns genomewide motif instances.

The genome and build information should be provided and relevant BS genomes packages such as BSgenome.Mmusculus.UCSC.mm10 or BSgenome.Hsapiens.UCSC.hg38 must be installed for the used genome and builds.

Usage

```
findMotifs(
  motif,
  mismatchNumber,
  genome,
  genomeBuild,
  DB = "UCSC",
  mainCHRs = TRUE,
  firstCHR = FALSE,
  MotifLocationName = "Motif_Locations",
  limitedRegion = NA
)
```

Arguments

motif	motif characters in nucleotide IUPAC format
mismatchNumber	Number of mismatch allowed to match with motif
genome	The genome name such as "Hsapiens", "Mmusculus", "Dmelanogaster"
genomeBuild	The genome build such as "hg38", "hg19", "mm10", "dm3"
DB	The database of genome build. default: "UCSC"
mainCHRs	If true only the major chromosome are considered, if FALSE Random, Uncharacterised, and Mithocondrial chromosomes are also considered
firstCHR	If true only Chr1 is used to find motifs. Default is FALSE
MotifLocationName	The name of the file of the motif locations
limitedRegion	If specified the motifs are detected in the provided granges

Value

No return value

---

fitKernelDensity *Fit a kernel density distribution to the obersever heuristic distribution*

Description

This function gets heuristic distribution of short reads around binding sites, total numer of short reads and window size as number of neucleotid around binding sites.

It fits a kernel to the distribution and return the distribution as output. The total sum of returned values is equal to one.

Usage

```
fitKernelDensity(heuristicDistribution, totalShortReads, windowSize)
```

Arguments

heuristicDistribution	Original short distribution
totalShortReads	Total number of short reads
windowSize	Window size around binding site. The total region would be 2*windowSize+1

Value

kernel returns fitted kernel distribution of short reads around binding sites

generate1ntBedAlignment

Convert bam and bed files to 1 nucleotide bed

Description

Take alignment files in bam or bed fomat and convert them to 1 nucleotide bed file

Usage

```
generate1ntBedAlignment(InputFile, bedFile, format = "")
```

Arguments

InputFile	Original alignment file name
bedFile	Name of output 1nt bed file
format	alignment format and should be one of these: "BAMPE", "BAMSE", "BEDPE", "BEDSE"

Value

No return value

Motif2Site

Detect and Recenter binding sites from ChIP-seq experiments

Description

Take ChIP-seq and motifs and detect Binding sites. It also combines/compares binding sites across experiments. Here is a synthetic example of differential Fur binding sites in E.coli:

Examples

```
# FUR candidate motifs in NC_000913 E. coli
FurMotifs=system.file("extdata", "FurMotifs.bed", package="Motif2Site")

# ChIP-seq datasets fe in bed single end format
IPFe <- c(system.file("extdata", "FUR_fe1.bed", package="Motif2Site"),
        system.file("extdata", "FUR_fe2.bed", package="Motif2Site"))
Inputs <- c(system.file("extdata", "Input1.bed", package="Motif2Site"),
            system.file("extdata", "Input2.bed", package="Motif2Site"))
FURfeBedInputStats <-
  DetectBindingSitesBed(BedFile=FurMotifs,
                        IPfiles=IPFe,
                        BackgroundFiles=Inputs,
                        genome="Ecoli",
                        genomeBuild="20080805",
                        DB="NCBI",
                        expName="FUR_Fe_BedInput",
                        format="BEDSE"
```

```
                        )

# ChIP-seq datasets dpd in bed single end format
IPDpd <- c(system.file("extdata", "FUR_dpd1.bed", package="Motif2Site"),
        system.file("extdata", "FUR_dpd2.bed", package="Motif2Site"))
FURdpdBedInputStats <-
  DetectBindingSitesBed(BedFile=FurMotifs,
                        IPfiles=IPDpd,
                        BackgroundFiles=Inputs,
                        genome="Ecoli",
                        genomeBuild="20080805",
                        DB="NCBI",
                        expName="FUR_Dpd_BedInput",
                        format="BEDSE"
                       )


# Combine all FUR binding sites into one table
corMAT <- recenterBindingSitesAcrossExperiments(
  expLocations=c("FUR_Fe_BedInput","FUR_Dpd_BedInput"),
  experimentNames=c("FUR_Fe","FUR_Dpd"),
  expName="combinedFUR",
  )
corMAT

# Differential binding sites across FUR conditions fe vs dpd
diffFUR <- pairwisDifferential(tableOfCountsDir="combinedFUR",
                               exp1="FUR_Fe",
                               exp2="FUR_Dpd",
                               FDRcutoff=0.05,
                               logFCcuttoff=1
                               )

FeUp <- diffFUR[[1]]
DpdUp <- diffFUR[[2]]
TotalComparison <- diffFUR[[3]]
head(TotalComparison)
```

`motifBindingNegativeBinomialCount` *Model IP and Input count values with negative Binomal*

Description

Using edgeR TMM normalization and estimating dispersion as well as Adapting exact test function from edgeR to model IP vs Input counts.

To make this function memory effcient motifs into smaller sets and compute them seperately and combine them at the end.

Usage

```
motifBindingNegativeBinomialCount(
  countTableFile,
```

```
  replicateNumber,
  outputFile,
  currentDir
)
```

Arguments

countTableFile	Table of counts which contains all IP and Input value raw counts
replicateNumber	experiment replicate number
outputFile	The name of the output file generated by this function
currentDir	Directory for I/O operations

Value

A dataframe includes fold enrichment, pvalue, and normalized count values

---

motifChipCount *count short reads related to each motif for a given ChIPseq file*

Description

count 1nt short reads related to each motif for a given ChIPseq file.

Usage

```
motifChipCount(motifFile, chipFile, windowSize, outputName)
```

Arguments

motifFile	File contains motifs
chipFile	ChIP-seq 1nt alignment locations in bed format
windowSize	Window size around binding site. The total region would be 2*windowSize+1
outputName	Name of the output table

Value

Total number of short reads in motif reagions

motifCount *count short reads around motifs for all ChIP-seq experiments*

Description

count short reads related to each motif for all ChIPseq files both IP and Input.

Usage

```
motifCount(motifFile, chipSeq, windowSize, outputName, currentDir)
```

Arguments

motifFile	File contains motifs
chipSeq	dataframe of ChIP-seq 1nt alignment location
windowSize	Window size around binding site. The total region would be 2*windowSize+1
outputName	Name of the output table
currentDir	Directory for I/O operations

Value

No return value

motifTablePreProcess *Process count data and perform negative binomial test*

Description

Remove unmmaped regions, low and high binding regions and regions without fold change, and call negative binomial or nb test for the remaining regions.

Usage

```
motifTablePreProcess(countTableFile, outFile, currentDir)
```

Arguments

countTableFile	Tabl of count values around motifs for all ChIP-seq experiments
outFile	The name of the output file
currentDir	Directory for I/O operations

Value

sequencingStatitics A dataframe consists of the ratio of non-sequenced, low-sequenced, ang high-sequenced regions.

NegativeBinomialTestWithReplicate *Negative binomial test of binding using all replicates*

Description

Adapted exact test function from edgeR to compare IP vs Input with replicates. Input is a DGELIST with common and tag-wise dispression has been already caluclated by edgeR commands.

It calculates abundaces with mglmOneGroup identical to edgeR. logFE was calculated identiacl to edgeR. For the pvalue test negative binomial test is performed on the calculated abundance.

Usage

```
NegativeBinomialTestWithReplicate(object, prior.count = 0.125)
```

Arguments

object	Table of counts which contains all IP and Input value counts, TMM normalized and contains dispersion values
prior.count	edgeR prior value

Value

log fold enrichment, pvalue, and normalized count values

pairwisDifferential *Detect differential motifs*

Description

Take combined matrix of motif counts generated by recenterBindingSitesAcrossExperiments, and experiment names. It detect differential motifs using edgeR TMM nomralizaiton with Generalized linear model

Usage

```
pairwisDifferential(
  tableOfCountsDir = "",
  exp1,
  exp2,
  FDRcutoff = 0.05,
  logFCcuttoff = 1
)
```

Arguments

tableOfCountsDir	Directory which conatins the combined motifs and ChIP-seq count file
exp1	Experiment name which will be compared in pairwise comparison
exp2	Experiment name which will be compared in pairwise comparison
FDRcutoff	FDR cutoff applies on pvalue distribution
logFCcuttoff	log fold change cutoff

Value

A list of differential motifs, motif1 and motif2 as well as a table of total motifs and log fold changes

See Also

recenterBindingSitesAcrossExperiments

Examples

```
# FUR candidate motifs in NC_000913 E. coli
FurMotifs=system.file("extdata", "FurMotifs.bed", package="Motif2Site")

# ChIP-seq datasets fe in bed single end format
IPFe <- c(system.file("extdata", "FUR_fe1.bed", package="Motif2Site"),
        system.file("extdata", "FUR_fe2.bed", package="Motif2Site"))
Inputs <- c(system.file("extdata", "Input1.bed", package="Motif2Site"),
            system.file("extdata", "Input2.bed", package="Motif2Site"))
FURfeBedInputStats <-
  DetectBindingSitesBed(BedFile=FurMotifs,
   IPfiles=IPFe,
   BackgroundFiles=Inputs,
   genome="Ecoli",
   genomeBuild="20080805",
   DB="NCBI",
   expName="FUR_Fe_BedInput",
   format="BEDSE"
   )

# ChIP-seq datasets dpd in bed single end format
IPDpd <- c(system.file("extdata", "FUR_dpd1.bed", package="Motif2Site"),
        system.file("extdata", "FUR_dpd2.bed", package="Motif2Site"))
FURdpdBedInputStats <-
  DetectBindingSitesBed(BedFile=FurMotifs,
   IPfiles=IPDpd,
   BackgroundFiles=Inputs,
   genome="Ecoli",
   genomeBuild="20080805",
   DB="NCBI",
   expName="FUR_Dpd_BedInput",
   format="BEDSE"
   )


# Combine all FUR binding sites into one table
corMAT <- recenterBindingSitesAcrossExperiments(
  expLocations=c("FUR_Fe_BedInput","FUR_Dpd_BedInput"),
```

```
  experimentNames=c("FUR_Fe","FUR_Dpd"),
  expName="combinedFUR",
  )

# Differential binding sites across FUR conditions fe vs dpd
diffFUR <- pairwisDifferential(tableOfCountsDir="combinedFUR",
   exp1="FUR_Fe",
   exp2="FUR_Dpd",
   FDRcutoff=0.05,
   logFCcuttoff=1
   )

FeUp <- diffFUR[[1]]
DpdUp <- diffFUR[[2]]
TotalComparison <- diffFUR[[3]]
head(TotalComparison)
```

`quiet` *Suppress messages generated by in external package*

Description

mixtools and MASS::fitdistr generates warning by cat which is suppressed by this funcitons

Usage

```
quiet(func)
```

Arguments

`func`	functional input call for which cat messages should be supressed

Value

No return value

`recenterBindingSitesAcrossExperiments` *Combine binding sites across experiments*

Description

Take experiment folder locations and experiment names and combine them into a combined matrix of motifs and ChIP-seq counts

Experiment folders must be generated either by `DetectBindingSitesBed` or `DetectBindingSitesMotif`.

Usage

```
recenterBindingSitesAcrossExperiments(
  expLocations,
  experimentNames,
  expName = "combinedData",
  fdrValue = 0.05,
  fdrCrossExp = 0.001
)
```

Arguments

expLocations	The path to the experiment folders
experimentNames	Name of the experiment to be used in combined ChIP-seq
expName	Name of the combined matrix
fdrValue	FDR cut-off to accept binding in each ChIP-seq experiments
fdrCrossExp	If no experiment fullfill this cutoff, the motif is not considered

Value

A pariwise Pearson correlation matrix across experiments

See Also

pairwisDifferential

Examples

```
# FUR candidate motifs in NC_000913 E. coli
FurMotifs=system.file("extdata", "FurMotifs.bed", package="Motif2Site")

# ChIP-seq datasets fe in bed single end format
IPFe <- c(system.file("extdata", "FUR_fe1.bed", package="Motif2Site"),
        system.file("extdata", "FUR_fe2.bed", package="Motif2Site"))
Inputs <- c(system.file("extdata", "Input1.bed", package="Motif2Site"),
            system.file("extdata", "Input2.bed", package="Motif2Site"))
FURfeBedInputStats <-
  DetectBindingSitesBed(BedFile=FurMotifs,
   IPfiles=IPFe,
   BackgroundFiles=Inputs,
   genome="Ecoli",
   genomeBuild="20080805",
   DB="NCBI",
   expName="FUR_Fe_BedInput",
   format="BEDSE"
   )

# ChIP-seq datasets dpd in bed single end format
IPDpd <- c(system.file("extdata", "FUR_dpd1.bed", package="Motif2Site"),
        system.file("extdata", "FUR_dpd2.bed", package="Motif2Site"))
FURdpdBedInputStats <-
  DetectBindingSitesBed(BedFile=FurMotifs,
   IPfiles=IPDpd,
   BackgroundFiles=Inputs,
```

```
   genome="Ecoli",
   genomeBuild="20080805",
   DB="NCBI",
   expName="FUR_Dpd_BedInput",
   format="BEDSE"
   )


# Combine all FUR binding sites into one table
corMAT <- recenterBindingSitesAcrossExperiments(
    expLocations=c("FUR_Fe_BedInput","FUR_Dpd_BedInput"),
    experimentNames=c("FUR_Fe","FUR_Dpd"),
    expName="combinedFUR",
    )
corMAT
```

`removeNonBellShapedMotifs` *Remove non-bell shpape motifs prior to binding signal decomposition*

Description

Gets motif locations and related short reads and returns the motifs which are non-skewed abs(skewness) < 0.3 and more short reads binds closer to site.

It counts around motif with interval windowSize and windowSize/2, if the smaller window is less than half of the larger one then motif is not considered as Bell-shape

Usage

```
removeNonBellShapedMotifs(motifLocations, readLocations, windowSize)
```

Arguments

`motifLocations`	A vector of motif locations
`readLocations`	A vector of 1nt short reads
`windowSize`	Window size around binding site. The total region would be 2*windowSize+1

Value

The coordinates of accepted motifs

`strongestMotif` *Returns the motif with the highest count*

Description

Gets motif locations and related short reads and returns the motif which include the highest number of short reads around it.

Usage

```
strongestMotif(motifLocations, readLocations, windowSize)
```

Arguments

`motifLocations`	A vector of motif locations
`readLocations`	A vector of 1nt short reads
`windowSize`	Window size around binding site. The total region would be 2*windowSize+1

Value

The strongest motif

Index