# Package 'scRepertoire'

October 14, 2021

Title A toolkit for single-cell immune receptor profiling

Version 1.2.0

#### **Description**

scRepertoire was built to process data derived from the 10x Genomics Chromium Immune Profiling for both T-cell receptor (TCR) and immunoglobulin (Ig) enrichment workflows and subsequently interacts with the popular Seurat and SingleCellExperiment R packages. It also allows for general analysis of single-cell clonotype information without the use of expression information. The package functions as a wrapper for Startrac and powerTCR R packages.

License Apache License 2.0

**Encoding UTF-8** 

LazyData true

RoxygenNote 7.1.1

**biocViews** Software, ImmunoOncology, SingleCell, Classification, Annotation, Sequencing

**Depends** ggplot2, R (>= 4.0)

**Imports** Biostrings, dplyr, reshape2, ggalluvial, stringr, vegan, powerTCR, SummarizedExperiment, plyr, parallel, doParallel, methods, utils, rlang

Suggests knitr, rmarkdown, BiocStyle, scater, circlize, scales, Seurat

VignetteBuilder knitr

git\_url https://git.bioconductor.org/packages/scRepertoire

git\_branch RELEASE\_3\_13

git\_last\_commit f61c711

git\_last\_commit\_date 2021-06-16

Date/Publication 2021-10-14

**Author** Nick Borcherding [aut, cre]

Maintainer Nick Borcherding <ncborch@gmail.com>

2 abundanceContig

# **R** topics documented:

abundanceContig	2
addVariable	3
alluvialClonotypes	4
clonalDiversity	5
clonalHomeostasis	6
clonalOverlap	7
clonalProportion	8
clonesizeDistribution	9
combineBCR	10
combineExpression	11
combineTCR	12
compareClonotypes	13
contig_list	14
expression2List	14
getCirclize	15
highlightClonotypes	16
lengthContig	17
occupiedscRepertoire	18
quantContig	
screp_example	
Startrac	
StartracDiversity	
stripBarcode	
subsetContig	
vizVgenes	23

abundanceContig

Demonstrate the relative abundance of clonotypes by group or sample.

24

## **Description**

Index

This function takes the output of combineTCR(), combineBCR(), or expression2List() and displays the number of clonotypes at specific frequencies by sample or group. Visualization can either be a line graph using calculated numbers or if scale = TRUE, the output will be a density plot. Multiple sequencing runs can be group together using the group parameter. If a matrix output for the data is preferred, set exportTable = TRUE.

# Usage

```
abundanceContig(
  df,
  cloneCall = "gene+nt",
  scale = FALSE,
  group = NULL,
```

addVariable 3

```
exportTable = FALSE
)
```

#### **Arguments**

df The product of combineTCR(), combineBCR(), or expression2List().

cloneCall How to call the clonotype - CDR3 gene (gene), CDR3 nucleotide (nt), CDR3

amino acid (aa), or C DR3 gene+nucleotide (gene+nt).

scale Converts the graphs into denisty plots in order to show relative distributions.

group The column header for which you would like to analyze the data.

exportTable Returns the data frame used for forming the graph to the visualization.

#### Value

ggplot of the total or relative adundance of clonotypes across quanta

#### **Examples**

```
#Making combined contig data
x <- contig_list
combined <- combineTCR(x, rep(c("PX", "PY", "PZ"), each=2),
rep(c("P", "T"), 3), cells ="T-AB")
abundanceContig(combined, cloneCall = "gene", scale = FALSE)</pre>
```

addVariable

Adding variables after the combination of contigs.

#### **Description**

This function adds variables to the product of combineTCR() combineBCR() or expression2List() to be used in later visualizations. For each element, the function will add a column (labled by name) with the variable. The length of the variable paramater needs to match the length of the combined object.

## Usage

```
addVariable(df, name = NULL, variables = NULL)
```

## **Arguments**

df The product of combineTCR() combineBCR() or expression2List().

name The column header to add.

variables The exact values to add to each element of the list.

4 alluvialClonotypes

#### Value

list of contigs with a new column (name).

## **Examples**

```
x <- contig_list
combined <- combineTCR(x, rep(c("PX", "PY", "PZ"), each=2),
rep(c("P", "T"), 3), cells ="T-AB")
combined <- addVariable(combined, name = "batch", variables = c(1,1,1,1,2,2))</pre>
```

alluvialClonotypes

Exploring interaction of clonotypes by seurat or SCE dynamics

#### **Description**

View the proportional contribution of clonotypes by seurat or SCE object meta data after combine-Expression(). The visualization is based on the ggalluvial package, which requires the aesthetics to be part of the axes that are visualized. Therefore, alpha, facet, and color should be part of the the axes you wish to view or will add an additional stratum/column to the end of the graph.

#### Usage

```
alluvialClonotypes(
   sc,
   cloneCall = c("gene", "nt", "aa", "gene+nt"),
   y.axes = NULL,
   color = NULL,
   alpha = NULL,
   facet = NULL
)
```

#### **Arguments**

SC	The seurat or SCE object to visualize after combineExpression(). For SCE objects, the cluster variable must be in the meta data under "cluster".
cloneCall	How to call the clonotype - CDR3 gene (gene), CDR3 nucleotide (nt) or CDR3 amino acid (aa), or CDR3 gene+nucleotide (gene+nt).
y.axes	The columns that will seperate the proportional visualizations.
color	The column header or clonotype(s) to be highlighted.
alpha	The column header to have gradieted opacity.
facet	The column label to seperate.

#### Value

Alluvial ggplot comparing clonotype distribution across selected parameters.

clonalDiversity 5

#### **Examples**

```
#Getting the combined contigs
combined <- combineTCR(contig_list, rep(c("PX", "PY", "PZ"), each=2),
rep(c("P", "T"), 3), cells ="T-AB")

#Getting a sample of a Seurat object
screp_example <- get(data("screp_example"))
sce <- suppressMessages(Seurat::UpdateSeuratObject(screp_example))
sce <- Seurat::as.SingleCellExperiment(sce)

#Using combineExpression()
sce <- combineExpression(combined, sce)

#Using alluvialClonotypes()
alluvialClonotypes(sce, cloneCall = "gene",
y.axes = c("Patient", "cluster"), color = "cluster")</pre>
```

clonalDiversity

Examine the clonal diversity of samples

## **Description**

This function calculates traditional measures of diversity - Shannon, inverse Simpson, Chao1 index, and abundance-based coverage estimators (ACE) by sample or group. The group paramter can be used to condense the individual samples. If a matrix output for the data is preferred, set exportTable = TRUE.

#### Usage

```
clonalDiversity(
  df,
  cloneCall = "gene+nt",
  group = "samples",
  exportTable = FALSE
)
```

#### **Arguments**

df The product of combineTCR(), combineBCR(), or expression2List().

cloneCall How to call the clonotype - CDR3 gene (gene), CDR3 nucleotide (nt) or CDR3

amino acid (aa), or CDR3 gene+nucleotide (gene+nt).

group The column header for which you would like to analyze the data.

exportTable Exports a table of the data into the global environment in addition to the visual-

ization

6 clonalHomeostasis

#### Value

ggplot of the diversity of clonotype sequences across list

#### **Examples**

```
#Making combined contig data
x <- contig_list
combined <- combineTCR(x, rep(c("PX", "PY", "PZ"), each=2),
rep(c("P", "T"), 3), cells ="T-AB")
clonalDiversity(combined, cloneCall = "gene")</pre>
```

clonalHomeostasis

Examining the clonal homeostasis

#### **Description**

This function calculates the space occupied by clonotype proportions. The grouping of these clonotypes is based on the parameter cloneTypes, at default, cloneTypes will group the clonotypes into bins of Rare = 0 to 0.0001, Small = 0.0001 to 0.001, etc. To adjust the proportions, change the number or labeling of the cloneTypes parameter. If a matrix output for the data is preferred, set exportTable = TRUE.

## Usage

#### Arguments

df The product of CombineContig() or expression2List() cloneTypes The cutpoints of the proportions.

cloneCall How to call the clonotype - CDR3 gene (gene), CDR3 nucleotide (nt) or CDR3

amino acid (aa), or CDR3 gene+nucleotide (gene+nt).

exportTable Exports a table of the data into the global environment in addition to the visual-

ization

#### Value

ggplot of the space occupied by the specific propotion of clonotypes

clonalOverlap 7

#### **Examples**

```
#Making combined contig data
x <- contig_list
combined <- combineTCR(x, rep(c("PX", "PY", "PZ"), each=2),
rep(c("P", "T"), 3), cells ="T-AB")
clonalHomeostasis(combined, cloneCall = "gene")</pre>
```

clonalOverlap

Examining the clonal overlap between groups or samples

## Description

This functions allows for the caclulation and visualizations of the overlap coefficient or morisita index for clonotypes using the product of combineTCR(), combineBCR() or expression2list(). The overlap coefficient is calculated using the intersection of clonotypes divided by the length of the smallest component. Morisita index is estimating the dispersion of a population, more information can be found [here](https://en.wikipedia.org/wiki/Morisita If a matrix output for the data is preferred, set exportTable = TRUE.

#### Usage

```
clonalOverlap(
   df,
   cloneCall = c("gene", "nt", "aa", "gene+nt"),
   method = c("overlap", "morisita"),
   exportTable = FALSE
)
```

## Arguments

df	$The \ product \ of \ combine TCR(), \ combine BCR(), \ or \ expression 2 List().$
cloneCall	How to call the clonotype - CDR3 gene (gene), CDR3 nucleotide (nt) or CDR3 amino acid (aa), or CDR3 gene+nucleotide (gene+nt).
method	The method to calculate the overlap, either the overlap coefficient or morisita index.
exportTable	Exports a table of the data into the global environment in addition to the visualization

#### Value

ggplot of the clonotypic overlap between elements of a list

8 clonalProportion

#### **Examples**

```
#Making combined contig data
x <- contig_list
combined <- combineTCR(x, rep(c("PX", "PY", "PZ"), each=2),
rep(c("P", "T"), 3), cells ="T-AB")
clonalOverlap(combined, cloneCall = "gene", method = "overlap")</pre>
```

clonalProportion

Examining the clonal space occupied by specific clonotypes

## **Description**

This function calculates the relative clonal space occupied by the clonotypes. The grouping of these clonotypes is based on the parameter split, at default, split will group the clonotypes into bins of 1:10, 11:100, 101:1001, etc. To adjust the clonotypes selected, change the numbers in the variable split. If a matrix output for the data is preferred, set exportTable = TRUE.

#### Usage

```
clonalProportion(
    df,
    split = c(10, 100, 1000, 10000, 30000, 1e+05),
    cloneCall = "gene+nt",
    exportTable = FALSE
)
```

## **Arguments**

df The product of combineTCR(), combineBCR(), or expression2List().

split The cutpoints for the specific clonotypes.

cloneCall How to call the clonotype - CDR3 gene (gene), CDR3 nucleotide (nt) or CDR3

amino acid (aa), or CDR3 gene+nucleotide (gene+nt).

exportTable Exports a table of the data into the global environment in addition to the visual-

ization

#### Value

ggplot of the space occupied by the specific rank of clonotypes

```
#Making combined contig data
x <- contig_list
combined <- combineTCR(x, rep(c("PX", "PY", "PZ"), each=2),
rep(c("P", "T"), 3), cells ="T-AB")
clonalProportion(combined, cloneCall = "gene")</pre>
```

clonesizeDistribution 9

clonesizeDistribution Hierarchical clustering of clonotypes on clonotype size and Jensen-Shannon divergence

## Description

This function produces a heirachial clustering of clonotypes by sample using the Jensen-Shannon distance and discrete gamma-GPD spliced threshold model in the [powerTCR R package] (https://bioconductor.org/packages/Please read and cite PMID: 30485278 if using the function for analyses. If a matrix output for the data is preferred set exportTable = TRUE.

## Usage

```
clonesizeDistribution(
  df,
  cloneCall = "gene+nt",
  method = "ward.D2",
  exportTable = FALSE
)
```

#### **Arguments**

df The product of combineTCR(), combineBCR(), or expression2List().

cloneCall How to call the clonotype - CDR3 gene (gene), CDR3 nucleotide (nt), CDR3

amino acid (aa), or CDR3 gene+nucleotide (gene+nt).

method The clustering paramater for the dendrogram.

exportTable Returns the data frame used for forming the graph.

#### Value

ggplot dendrogram of the clone size distribution

```
#Making combined contig data
x <- contig_list
combined <- combineTCR(x, rep(c("PX", "PY", "PZ"), each=2),
rep(c("P", "T"), 3), cells ="T-AB")
clonesizeDistribution(combined, cloneCall = "gene+nt", method="ward.D2")</pre>
```

10 combineBCR

combineBCR

Combining the list of B Cell Receptor contigs

#### **Description**

This function consolidates a list of BCR sequencing results to the level of the individual cell barcodes. Using the samples and ID parameters, the function will add the strings as prefixes to prevent issues with repeated barcodes. The resulting new barcodes will need to match the seurat or SCE object in order to use, combineExpression. Unlike combineTCR(), combineBCR() produces a column CTstrict of an index of nucleotide sequence and the corresponding v-gene. This index automatically calculates the Hammings distance between sequences of the same length and will index sequences with > 0.85 normalized Hammings distance with the same ID. If nucleotide sequences meet the threshold, ":HD" will be added to the CTstrict column string.

#### Usage

```
combineBCR(
  df,
  samples = NULL,
  ID = NULL,
  removeNA = FALSE,
  removeMulti = FALSE)
```

#### **Arguments**

df List of filtered contig annotations from 10x Genomics.

samples The labels of samples.

The additional sample labeling option.

removeNA This will remove any chain without values.

removeMulti This will remove barcodes with greater than 2 chains.

#### Value

List of clonotypes for individual cell barcodes

```
#Data derived from the 10x Genomics intratumoral NSCLC B cells
BCR <- read.csv("https://ncborcherding.github.io/vignettes/b_contigs.csv",
stringsAsFactors = FALSE)
combined <- combineBCR(BCR, samples = "Patient1", ID = "Time1")</pre>
```

combineExpression 11

combineExpression

Adding clonotype information to a seurat or SCE object

## **Description**

This function adds the immune receptor information to the seurat or SCE object to the meta data. By defualt this function also calculates the frequencies of the clonotypes by sequencing run (groupBy = "none"). To change how the frequencies are calculated, select a column header for the groupBy variable. Importantly, before using combineExpression() ensure the barcodes of the seurat or SCE object match the barcodes in the output of the combinedContig() call. Check changeNames() to change the prefix of the seurat object. If the dominant clonotypes have a greater frequency than 500, adjust the cloneTypes variable.

## Usage

```
combineExpression(
   df,
   sc,
   cloneCall = "gene+nt",
   groupBy = "none",
   cloneTypes = c(None = 0, Single = 1, Small = 5, Medium = 20, Large = 100,
        Hyperexpanded = 500),
   filterNA = FALSE
)
```

#### **Arguments**

df	The product of CombineTCR() or CombineBCR().
sc	The seurat or SingleCellExperiment (SCE) object to attach
cloneCall	How to call the clonotype - CDR3 gene (gene), CDR3 nucleotide (nt) CDR3 amino acid (aa), or CDR3 gene+nucleotide (gene+nt).
groupBy	The column label in the combined contig object in which clonotype frequency will be calculated.
cloneTypes	The bins for the grouping based on frequency
filterNA	Method to subset seurat object of barcodes without clonotype information

#### Value

seurat or SingleCellExperiment object with attached clonotype information

```
#Getting the combined contigs
combined <- combineTCR(contig_list, rep(c("PX", "PY", "PZ"), each=2),
rep(c("P", "T"), 3), cells ="T-AB")</pre>
```

12 combineTCR

```
#Getting a sample of a Seurat object
screp_example <- get(data("screp_example"))
sce <- suppressMessages(Seurat::UpdateSeuratObject(screp_example))
sce <- Seurat::as.SingleCellExperiment(sce)

#Using combineExpression()
sce <- combineExpression(combined, sce)</pre>
```

combineTCR

Combining the list of T Cell Receptor contigs

#### **Description**

This function consolidates a list of TCR sequencing results to the level of the individual cell barcodes. Using the samples and ID parameters, the function will add the strings as prefixes to prevent issues with repeated barcodes. The resulting new barcodes will need to match the seurat or SCE object in order to use, combineExpression. Several levels of filtering exist - remove or filterMulti are parameters that control how the function deals with barcodes with multiple chains recovered.

## Usage

```
combineTCR(
  df,
  samples = NULL,
  ID = NULL,
  cells = c("T-AB", "T-GD"),
  removeNA = FALSE,
  removeMulti = FALSE,
  filterMulti = FALSE
)
```

## **Arguments**

df List of filtered contig annotations from 10x Genomics.

samples The labels of samples.

The additional sample labeling option.

The type of T cell - T cell-AB or T cell-GD

removeNA

This will remove any chain without values.

removeMulti This will remove barcodes with greater than 2 chains.

filterMulti This option will allow for the selection of the 2 corresponding chains with the

highest expression for a single barcode.

#### Value

List of clonotypes for individual cell barcodes

compareClonotypes 13

#### **Examples**

```
combineTCR(contig_list, rep(c("PX", "PY", "PZ"), each=2),
rep(c("P", "T"), 3), cells ="T-AB")
```

compareClonotypes

Demonstrate the difference in clonal proportion between clonotypes

## Description

This function produces an alluvial or area graph of the proportion of the indicated clonotypes for all or selected samples. Clonotypes can be selected using the clonotypes parameter with the specific sequence of interest or using the number parameter with the top n clonotypes by proportion to be visualized. If multiple clonotypes have the same proportion and are within the selection by the number parameter, all the clonotypes will be visualized. In this instance, if less clonotypes are desired, reduce the number parameter.

#### Usage

```
compareClonotypes(
  df,
  cloneCall = "gene+nt",
  samples = NULL,
  clonotypes = NULL,
  numbers = NULL,
  graph = "alluvial",
  exportTable = FALSE
)
```

## Arguments

df	The product of combineTCR(), combineBCR(), or expression2List()	
cloneCall	How to call the clonotype - CDR3 gene (gene), CDR3 nucleotide (nt), CI amino acid (aa), or CDR3 gene+nucleotide (gene+nt).	
samples	The specific samples to isolate for visualization.	
clonotypes	The specific sequences of interest.	
numbers	The top number clonotype sequences.	
graph	The type of graph produced, either "alluvial" or "area".	
exportTable	Returns the data frame used for forming the graph.	

#### Value

ggplot of the proportion of total sequencing read of selecting clonotypes

14 expression2List

#### **Examples**

```
#Making combined contig data
x <- contig_list
combined <- combineTCR(x, rep(c("PX", "PY", "PZ"), each=2),
rep(c("P", "T"), 3), cells ="T-AB")
compareClonotypes(combined, numbers = 10,
samples = c("PX_P", "PX_T"), cloneCall="aa")</pre>
```

contig\_list

A data set of T cell contigs as a list outputed from the filter\_contig\_annotation files.

#### Description

A data set of T cell contigs as a list outputed from the filter\_contig\_annotation files.

expression2List

Allows users to take the meta data in seurat/SCE and place it into a list that will work with all the functions

#### Description

Allows users to perform more fundamental measures of clonotype analysis using the meta data from the seurat or SCE object. For Seurat objects the active identity is automatically added as "cluster". Reamining grouping parameters or SCE or Seurat objects must appear in the meta data.

#### Usage

```
expression2List(sc, group)
```

#### **Arguments**

sc object after combineExpression().

group The column header to group the new list by

#### Value

list derived from the meta data of single-cell object with elements divided by the group parameter

getCirclize 15

#### **Examples**

```
#Getting the combined contigs
combined <- combineTCR(contig_list, rep(c("PX", "PY", "PZ"), each=2),
rep(c("P", "T"), 3), cells ="T-AB")

#Getting a sample of a Seurat object
screp_example <- get(data("screp_example"))
sce <- suppressMessages(Seurat::UpdateSeuratObject(screp_example))
sce <- Seurat::as.SingleCellExperiment(sce)

#Using expression2List
newList <- expression2List(sce, group = "seurat_clusters")</pre>
```

getCirclize

Generate data frame to be used with circlize R package to visualize clonotypes as a chord diagram.

## **Description**

This function will take the meta data from the product of combineExpression() and generate a relational data frame to be used for a chord diagram. The output is a measure of relative clonotype overlap between groups and does not reflect exact clonotype matches between groups.

#### Usage

```
getCirclize(sc, cloneCall = "gene+nt", groupBy = NULL, proportion = FALSE)
```

## **Arguments**

sc object after combineExpression().

cloneCall How to call the clonotype - CDR3 nucleotide (nt), CDR3 amino acid (aa).

groupBy The group header for which you would like to analyze the data.

proportion Binary will calculate relationship as unique clonotypes (proportion = TRUE) or

proportion of unique clonotypes (proportion = FALSE)

## Value

data frame of shared clonotypes between groups

#### Author(s)

Dillon Corvino, Nick Borcherding

#### **Examples**

```
#Getting the combined contigs
combined <- combineTCR(contig_list, rep(c("PX", "PY", "PZ"), each=2),
rep(c("P", "T"), 3), cells ="T-AB")

#Getting a sample of a Seurat object
screp_example <- get(data("screp_example"))
screp_example <- combineExpression(combined, screp_example)

#Getting data frame output for Circilize
circles <- getCirclize(screp_example, groupBy = "seurat_clusters")</pre>
```

highlightClonotypes

Highlighting specific clonotypes in Seurat

#### **Description**

Use a specific clonotype sequence to highlight on top of the dimensional reduction in seurat object.

#### Usage

```
highlightClonotypes(
   sc,
   cloneCall = c("gene", "nt", "aa", "gene+nt"),
   sequence = NULL
)
```

#### **Arguments**

sc The seurat object to attach

cloneCall How to call the clonotype - CDR3 gene (gene), CDR3 nucleotide (nt), CDR3

amino acid (aa), or CDR3 gene+nucleotide (gene+nt).

sequence The specifc sequence or sequence to highlight

## Value

DimPlot with highlighted clonotypes

```
#' #Getting the combined contigs
combined <- combineTCR(contig_list, rep(c("PX", "PY", "PZ"), each=2),
rep(c("P", "T"), 3), cells ="T-AB")

#Getting a sample of a Seurat object
screp_example <- get(data("screp_example"))</pre>
```

lengthContig 17

```
#Using combineExpresion()
screp_example <- combineExpression(combined, screp_example )
#Using highlightClonotype()
screp_example <- highlightClonotypes(screp_example, cloneCall= "aa",
sequence = c("CAVNGGSQGNLIF_CSAEREDTDTQYF"))</pre>
```

lengthContig

Demonstrate the distribution of lengths filtered contigs.

## **Description**

This function takes the output of combineTCR(), combineBCR(), or expression2List() and displays either the nucleotide (nt) or amino acid (aa) sequence length. The sequence length visualized can be selected using the chains parameter, either the combined clonotype (both chains) or across all single chains. Visualization can either be a histogram or if scale = TRUE, the output will be a density plot. Multiple sequencing runs can be group together using the group parameter. If a matrix output for the data is preferred, set exportTable = TRUE.

## Usage

```
lengthContig(
   df,
   cloneCall = "aa",
   group = NULL,
   scale = FALSE,
   chains = "combined",
   exportTable = FALSE
)
```

## Arguments

cloneCall How to call the clonotype - CDR3 nucleotide (nt), CDR3 amino acid (aa).  group The group header for which you would like to analyze the data.  scale Converts the graphs into denisty plots in order to show relative distributions.  chains Whether to keep clonotypes "combined" or visualize by chain.  exportTable Returns the data frame used for forming the graph.	df	The product of combineTCR(), combineBCR(), or expression2List()	
scale Converts the graphs into denisty plots in order to show relative distributions.  chains Whether to keep clonotypes "combined" or visualize by chain.	cloneCall	How to call the clonotype - CDR3 nucleotide (nt), CDR3 amino acid (aa).	
chains Whether to keep clonotypes "combined" or visualize by chain.	group	The group header for which you would like to analyze the data.	
1 71	scale	Converts the graphs into denisty plots in order to show relative distributions.	
exportTable Returns the data frame used for forming the graph.	chains	Whether to keep clonotypes "combined" or visualize by chain.	
	exportTable	Returns the data frame used for forming the graph.	

#### Value

ggplot of the discrete or relative length distributions of clonotype sequences

#### **Examples**

```
#Making combined contig data
x <- contig_list
combined <- combineTCR(x, rep(c("PX", "PY", "PZ"), each=2),
rep(c("P", "T"), 3), cells ="T-AB")
lengthContig(combined, cloneCall="aa", chains = "combined")</pre>
```

occupiedscRepertoire Visualize the number of single cells with clonotype frequencies by cluster

#### **Description**

View the count of clonotypes frequency group in seurat or SCE object meta data after combineExpression(). The visualization will take the new meta data variable "cloneType" and plot the number of cells with each designation using a secondary variable, like cluster. Credit to the idea goes to Dr. Carmonia and his work with [ProjectTIL](https://github.com/carmonalab/ProjecTILs).

## Usage

```
occupiedscRepertoire(sc, x.axis = "cluster", exportTable = FALSE)
```

## Arguments

sc	The seurat or SCE object to visualize after combineExpression(). For SCE objects, the cluster variable must be in the meta data under "cluster".
x.axis	The variable in the meta data to graph along the x.axis
exportTable	Exports a table of the data into the global environment in addition to the visualization

#### Value

Stacked bar plot of counts of cells by clonotype frequency group

```
#Getting the combined contigs
combined <- combineTCR(contig_list, rep(c("PX", "PY", "PZ"), each=2),
rep(c("P", "T"), 3), cells ="T-AB")

#Getting a sample of a Seurat object
screp_example <- get(data("screp_example"))
sce <- suppressMessages(Seurat::UpdateSeuratObject(screp_example))
sce <- Seurat::as.SingleCellExperiment(sce)

#Using combineExpression()
sce <- combineExpression(combined, sce)</pre>
```

quantContig 19

```
#Using occupiedscRepertoire()
occupiedscRepertoire(sce, x.axis = "cluster")
table <- occupiedscRepertoire(sce, x.axis = "cluster", exportTable = TRUE)</pre>
```

quantContig

Quantify the unique clonotypes in the filtered contigs.

#### **Description**

This function takes the output from combineTCR(), combineBCR(), or expression2List() and quantifies unique clonotypes. The unique clonotypes can be either reported as a raw output or scaled to the total number of clonotypes recovered using the scale parameter. Multiple sequencing runs can be group together using the group parameter. If a matrix output for the data is preferred, set exportTable = TRUE.

#### Usage

```
quantContig(
   df,
   cloneCall = "gene+nt",
   scale = FALSE,
   group = NULL,
   exportTable = FALSE
)
```

#### **Arguments**

df The product of combineTCR() combineBCR() or expression2List().

cloneCall How to call the clonotype - CDR3 gene (gene), CDR3 nucleotide (nt), CDR3

amino acid (aa), or CDR3 gene+nucleotide (gene+nt).

scale Converts the graphs into percentage of unique clonotypes.

group The column header used for grouping.

exportTable Returns the data frame used for forming the graph

## Value

ggplot of the total or relative unique clonotypes

```
#Making combined contig data
x <- contig_list
combined <- combineTCR(x, rep(c("PX", "PY", "PZ"), each=2),
rep(c("P", "T"), 3), cells ="T-AB")
quantContig(combined, cloneCall="gene+nt", scale = TRUE)</pre>
```

20 Startrac

screp_example	A seurat object of 1000 single T cells derived from 3 clear cell renal carcinoma patients.

#### Description

A seurat object of 1000 single T cells derived from 3 clear cell renal carcinoma patients.

Startrac The Startrac Class	Startrac The Startrac Class
-----------------------------	-----------------------------

#### **Description**

The Startrac object store the data for tcr-based T cell dynamics analyis. The slots contained in Startrac object are listed below:

#### **Slots**

- aid character. aid of the object, used for identification of the object. For example, patient id. default: "AID"
- cell.data data.frame. Each line for a cell, and these columns as required: 'Cell\_Name', 'clone.id', 'patient', 'majorCluster', 'loc'
- cell.perm.data object. list of 'Startrac" objects constructed from permutated cell data
- clonotype.data data.frame. Each line for a clonotype; contain the clonotype level indexes information
- cluster.data data.frame. Each line for a cluster; contain the cluster level indexes information
- pIndex.migr data.frame. Each line for a cluster; pairwise migration index between the two locations indicated in the column name.
- pIndex.tran data.frame. Each line for a cluster; pairwise transition index between the two major clusters indicated by the row name and column name.
- cluster.sig.data data.frame. Each line for a cluster; contains the p values of cluster indices.
- pIndex.sig.migr data.frame. Each line for a cluster; contains the p values of pairwise migration indices.
- pIndex.sig.tran data.frame. Each line for a cluster; contains the p values of pairwise transition indices.
- clonotype.dist.loc matrix. Each line for a clonotype and describe the cells distribution among the locations.
- clonotype.dist.cluster matrix. Each line for a clonotype and describe the cells distribution among the clusters.
- clust.size array. Number of cells of each major cluster.
- patient.size array. Number of cells of each patient.
- clone.size array. Number of cells of each clone.
- clone2patient array. Mapping from patient id to clone id.

StartracDiversity 21

rtracDiversity Diversity indices for single-cell RNA-seq
--

## Description

This function utilizes the Startrac R package derived from [PMID: 30479382](https://pubmed.ncbi.nlm.nih.gov/30479382/) Required to run the function, the "type" variable needs to include the difference in where the cells were derived. The output of this function will produce 3 indices: expa (clonal expansion), migra (cross-tissue migration), and trans (state transition). In order to understand the underlying analyses of the outputs please read and cite the linked manuscript.

## Usage

```
StartracDiversity(
    sc,
    type = "Type",
    sample = NULL,
    by = "overall",
    exportTable = FALSE
)
```

## Arguments

sc	The seurat or SCE object to visualize after combineExpression(). For SCE objects, the cluster variable must be in the meta data under "cluster".
type	The column header in the meta data that gives the where the cells were derived from, not the patient sample IDs
sample	The column header corresponding to individual samples or patients.
by	Method to subset the indices by either overall (across all samples) or by specific group
exportTable	Returns the data frame used for forming the graph

## Value

ggplot object of Startrac diversity metrics

## Description

Removing any additional prefixes to the barcodes of filtered contigs.

22 subsetContig

#### Usage

```
stripBarcode(contigs, column = 1, connector = "_", num_connects = 3)
```

## **Arguments**

contigs The raw loaded filtered\_contig\_annotation.csv column The column in which the barcodes are listed

connector The type of character in which is attaching the defualt barcode with any other

characters

num\_connects The number of strings combined with the connectors

#### Value

list with the suffixes of the barcodes removed.

#### **Examples**

```
stripBarcode(contig_list[[1]], column = 1, connector = "_", num_connects = 1)
```

subsetContig	Subset the product of combineTCR() combineBCR() or expres-
	sion2List()

#### **Description**

This function allows for the subsetting of the product of combineTCR() combineBCR() or expression2List() by the name of the individual list element. In general the names of are samples + \_ + ID, allowing for users to subset the product of combineTCR(), combineBCR(), or expression2List() across a string or individual name.

## Usage

```
subsetContig(df, name, variables = NULL)
```

#### **Arguments**

df The product of combineTCR(), combineBCR(), or expression2List().

name The column header you'd like to use to subset.

variables The values to subset by, must be in the names(df).

#### Value

list of contigs that have been filtered for the name parameter

vizVgenes 23

#### **Examples**

```
x <- contig_list
combined <- combineTCR(x, rep(c("PX", "PY", "PZ"), each=2),
rep(c("P", "T"), 3), cells ="T-AB")
subset <- subsetContig(combined, name = "sample", variables = c("PX"))</pre>
```

vizVgenes

Visualizing the distribution of TCR V gene usage

## Description

This function will allow for the visualizing the distribution of the V-genes of the TCR by categorical variables.

## Usage

```
vizVgenes(
  df,
  TCR = "TCR1",
  facet.x = "sample",
  facet.y = NULL,
  fill = NULL,
  exportTable = FALSE
)
```

#### **Arguments**

df	The product of combineTCR(), combineBCR(), or expression2List().
TCR	Which TCR chain to use, TCR1 = TCRA or TCR2 = TCRB
facet.x	Categorical variable which to seperate by along x-axis
facet.y	Categorical variable which to seperate by along y-axis
fill	Categorical variable which to add color to bar chart
exportTable	Returns the data frame used for forming the graph.

#### Value

ggplot bar diagram of vgene counts

```
#Making combined contig data
x <- contig_list
combined <- combineTCR(x, rep(c("PX", "PY", "PZ"), each=2),
rep(c("P", "T"), 3), cells ="T-AB")
vizVgenes(combined, TCR = "TCR1", facet.x = "sample")</pre>
```

# **Index**

```
abundance {\tt Contig}, {\tt 2}
addVariable, 3
{\it alluvialClonotypes, 4}
clonalDiversity, 5
clonalHomeostasis, 6
clonalOverlap, 7
clonalProportion, 8
{\tt clonesizeDistribution}, 9
combineBCR, 10
combineExpression, 10, 11, 12
combineTCR, 12
compareClonotypes, 13
contig_list, 14
{\tt expression2List}, \\ 14
getCirclize, 15
highlightClonotypes, 16
lengthContig, 17
occupiedscRepertoire, 18
{\tt quantContig}, {\tt 19}
screp_example, 20
Startrac, 20
Startrac-class (Startrac), 20
StartracDiversity, 21
stripBarcode, 21
subsetContig, 22
vizVgenes, 23
```