# Package 'redeemR'

June 3, 2023

Title R package for Regulatory multi-omics with Deep Mitochondrial mutation profiling

Version 1.0

**Description** Introduce a new approach for single-cell Regulatory multi-omics (transcriptomics and chromatin accessibility) with Deep Mitochondrial mutation profiling (~10-fold increase in detection rate), or ReDeeM. redeemR is the R package that facilitates mutation refining, lineage tracing, as well multiomics integration analysis.

```
License MIT LICENSE
Encoding UTF-8
Roxygen list(markdown = TRUE)
RoxygenNote 7.1.2
Imports ape,
      doMC,
      doParallel,
      dplyr,
      foreach,
      ggExtra,
      ggnewscale,
      ggplot2,
      ggtree,
      ggtreeExtra,
      gridExtra,
      igraph,
      Matrix,
      phangorn,
      phytools,
      pryr,
      qvalue,
      RColorBrewer,
      reshape2,
      SCAVENGE (>= 1.0.1),
      Seurat,
      stats,
      tibble,
      tidytree,
      treeio
```

Remotes github::sankaranlab/SCAVENGE

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# $\mathsf{R}$ topics documented:

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 ${\tt AddDatatoplot\_clustering}$ 

 $Add Data top lot\_clustering\ This\ prepare\ the\ clonal\ clustering\ data\ to\ plot$ 

# Description

AddDatatoplot\_clustering This prepare the clonal clustering data to plot

# Usage

```
AddDatatoplot_clustering(object, ...)
```

# **Arguments**

object redeemR class

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# Description

AddDatatoplot\_clustering This prepare the clonal clustering data to plot

# Usage

```
## S4 method for signature 'redeemR'
AddDatatoplot_clustering(object)
```

# Arguments

object mitoTracin class

#### Value

redeemR class

AddDist

AddDist This add Jaccard, Dice, Jaccard3W distance and stored in DistObjects

# Description

AddDist This add Jaccard, Dice, Jaccard3W distance and stored in DistObjects

# Usage

```
AddDist(object, ...)
```

# Arguments

object redeemR class

```
AddDist, redeemR-method
```

AddDist This add Jaccard, Dice, Jaccard3W distance and stored in DistObjects

# Description

AddDist This add Jaccard, Dice, Jaccard3W distance and stored in DistObjects

# Usage

```
## S4 method for signature 'redeemR'
AddDist(
  object,
  jaccard = T,
  dice = T,
  jaccard3w = T,
  w_jaccard = T,
  w_cosine = T,
  weightDF = NULL,
  NN = 1,
  LSIdist = T,
  dim = 2:50
)
```

# Arguments

object	mitoTracin class
jaccard	default=T
dice	default=T
jaccard3w	default=T
w_jaccard	default=T
w_cosine	default=T
NN	To replace NA, which means a variant shown in the object is not shown in the weight vector, with a number, default is 1 for jaccard system.
LSIdist	default=T
dim	the dimensions to use to calculate LSI distance default is 2:50
weight	A two column dataframe, "Variant"(The variant name should match cell-variant matrix column, e.g, Variants310TC), "weight" (numeric)

### Value

redeemR class

6 AddTree

AddHemSignature

Function to add hematopoietic signatures from Griffin\_Signatures

# Description

This function allows you to input a seurat object, add the signatures and return an seurat object

# Usage

```
AddHemSignature(object = Donor01_BMMC_Multiome_wrapper.filtered)
```

# Arguments

object

a seurat object

#### Value

a seurat object

AddTree

Add\_Tree Optional, if a phylogentic tree object phylo is already available, can be directly added to the redeemR

# Description

Add\_Tree Optional, if a phylogentic tree object phylo is already available, can be directly added to the redeemR

# Usage

```
AddTree(object, phylo, ...)
```

# Arguments

object redeemR class

phylo phyogenetic tree object

```
AddTree, redeemR-method
```

Add\_Tree Optional, if a phylogentic tree object phylo is already available, can be directly added to the redeemR class in slot TREE

# Description

Add\_Tree Optional, if a phylogentic tree object phylo is already available, can be directly added to the redeemR class in slot TREE

### Usage

```
## S4 method for signature 'redeemR'
AddTree(object, phylo, record = "")
```

# **Arguments**

object mitoTracin class

phylo phyogenetic tree object

#### Value

redeemR class

```
Add_AssignVariant Add_AssignVariant a function to assign variants to edges based on maximum likihood
```

### **Description**

Add\_AssignVariant a function to assign variants to edges based on maximum likihood

### Usage

```
Add_AssignVariant(redeemR, n.cores, ...)
```

# Arguments

```
object redeemR class
QualifiedTotalCts
    a big source data, usually at XXX/mitoV/final
```

8 Add\_DepthMatrix

```
{\tt Add\_AssignVariant, redeemR-method} \\ a \textit{ function to assign variants to edges based on maximum likihood}
```

### **Description**

a function to assign variants to edges based on maximum likihood

### Usage

```
## S4 method for signature 'redeemR'
Add_AssignVariant(redeemR = DN1_HSC_redeemR.VerySensitive, n.cores = 4)
```

### **Arguments**

redeemR

 $Need to have \ redeem R@Ctx.Mtx.depth \ (By\ Add\_DepthMatrix), \ redeem R@Cts.Mtx \ redeem R@Cts.Mtx.bi, \ redeem R@TREE$ 

#### Value

redeemR with @AssignedVarian list of two p is a probability matrix of variants vs edges (Rowsum is 1) and Variant.assign.report, a dataframe (VariantlEdge.Assignlprob)

Add\_DepthMatrix

Add\_DepthMatrix Optional, add a matrix with same dimension with the Cts.Mtx and Cts.Mtx.bi, which display the depths

### **Description**

Add\_DepthMatrix Optional, add a matrix with same dimension with the Cts.Mtx and Cts.Mtx.bi, which display the depths

### Usage

```
Add_DepthMatrix(object, QualifiedTotalCts, ...)
```

# Arguments

```
object redeemR class
QualifiedTotalCts
    a big source data, usually at XXX/mitoV/final
```

```
Add_DepthMatrix, redeemR-method
```

Add\_DepthMatrix Optional, add a matrix with same dimension with the Cts.Mtx and Cts.Mtx.bi, which display the depths

# Description

Add\_DepthMatrix Optional, add a matrix with same dimension with the Cts.Mtx and Cts.Mtx.bi, which display the depths

# Usage

```
## S4 method for signature 'redeemR'
Add_DepthMatrix(object)
```

### **Arguments**

```
object mitoTracin class

QualifiedTotalCts
    a big source data, usually at XXX/mitoV/final, If needed, edit V1, the cell name, which may have additional postfix due to combine
```

#### Value

redeemR class

```
add_derived_profile_info

This is a convinience function, internal borrowed from 
https://github.com/NickWilliamsSanger/treemut/blob/main/R/treemut.R#L68
```

# Description

This is a convinience function, internal borrowed from https://github.com/NickWilliamsSanger/treemut/blob/main/R/tree

### Usage

```
add_derived_profile_info(
  profile_df,
  samples = sprintf("s%s", 0:(nchar(profile_df$profile[1]) - 1))
)
```

Add\_tree\_cut a function to cut tree using assigned variant as branchlength on edge

# Description

Add\_tree\_cut a function to cut tree using assigned variant as branch-length on edge

#### Usage

```
Add_tree_cut(redeemR, MinCell, N, ...)
```

### **Arguments**

redeemR Need to have had the tree built

MinCell The minimum number of cells in each clone, otherwise merge with sibling

N branch length to cut the tree

```
Add_tree_cut, redeemR-method
```

a function to cut tree using assigned variant as branch-length on edge

### **Description**

a function to cut tree using assigned variant as branch-length on edge

#### Usage

```
## S4 method for signature 'redeemR'
Add_tree_cut(
   redeemR = DN4_stemcell_redeemR.seed.verysensitive,
   MinCell = 30,
   N = 1,
   prob.cut = 0.3,
   Dumpcut = 100
)
```

#### **Arguments**

redeemR Need to have had the tree built

MinCell The minimum number of cells in each clone, otherwise merge with sibling

N branch length to cut the tree

Dumpcut Number of can be tolerated to be removed to fulfill the right side. The small value-> Less unassignment, big clones

ATAC\_Wrapper 11

ATAC\_Wrapper

Wrap Seurat ATAC clustering

### **Description**

This function allows you to perform standard sc-ATAC clustering

### Usage

```
ATAC_Wrapper(MTX, res = 0.3, dim1 = 1, dim2 = 20)
```

### **Arguments**

MTX sparse Matrix of class "dgCMatrix", each row is a peak, each column is a cell,

res clustering resolution, default=0.5

#### Value

this returns seurat object with ATAC clustering

### **Examples**

bmmc.filtered.atac<-SeuratLSIClustering(PeakVSCell.filtered.Mtx) #each row is a peak, each

BinaryDist

Compute distances for binary distances

### **Description**

Compute distances for binary distances

# Usage

```
BinaryDist(M, method = "Jaccard")
```

### **Arguments**

M the binary matrix, Each row is a cell, each column is a variant, generated by

Make\_matrix

method distance method, choose from Jaccard, Dice, 3WJaccard, Simpson, Kulczyn-

ski2, Ochiai, Hamming

#### Value

dist object

### **Examples**

```
d.Jaccard<-BinaryDist(object@Cts.Mtx.bi,method="Jaccard")</pre>
```

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Clone\_FinderMarker Define a function to perform Find marker for top vs bottom clones This function was developed based on DN4T2.basics.ipynb

### **Description**

Define a function to perform Find marker for top vs bottom clones This function was developed based on DN4T2.basics.ipynb

#### Usage

```
Clone_FinderMarker(
  topClones,
  bottomClones,
  HSC_Multiome_wrapper = Donor04_HSC_Multiome_wrapper,
  HSC_redeemR,
  assay = "SCT",
  test = "wilcox"
)
```

#### **Arguments**

a vector of clone ID eg. c("1","3","7"),this must be in HSC\_redeemR@CellMeta\$Clone\_merge
bottomClones a vector of clone ID eg. c("2","5"), this must be in HSC\_redeemR@CellMeta\$Clone\_merge

HSC\_redeemR object for HSC

test the statistic method to use for DE, a wrapper function from Seurat FindAllMarkers

ob Seurat object (Multiomics), the postfix needs to be compatible with HSC\_redeemR,
the cells will be matched by cell names

ComputeRejectRate Function to compute the reject rate(The filtering rate in concensus variant calling)

# Description

This function allows you to computae the filtering rate for each single cell

### Usage

```
ComputeRejectRate(ob)
```

#### **Arguments**

ob The redeemR object

### Value

a modified ob with RejectRate added to @CellMeta

CountVperCell 13

CountVperCell

Internal function in plot\_variant

# **Description**

Internal function in plot\_variant

# Usage

```
CountVperCell(x, name, CellN)
```

### **Arguments**

```
x CellVar.Sum$VN
name c
CellN nrow(CellVar.Sum)
```

# **Examples**

```
CountVperCell(CellVar.Sum$VN,c,CellN=nrow(CellVar.Sum))
```

```
Create_mitoTracing Create_mitoTracing
```

# **Description**

This function is to create redeemR with basic information

# Usage

```
Create_mitoTracing(
  GTsummary_list,
  depth_list,
  feature.list_list,
  meta_list,
  labels,
  thr = "VerySensitive",
  qualifiedCellCut = 10,
  OnlyHetero = T,
  VAFcut = 1,
  Cellcut = 2,
  maxctscut = 2
)
```

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#### **Arguments**

GTsummary\_list

simply put GTSummary (Generated by CW\_mgatk.read) into list, this allows

mergeing multiple dataset this way.

depth\_list simply put depth(Generated by DepthSummary) into list, this allows mergeing

multiple dataset this way.

feature.list\_list

simply put feature.list(Generated by Vfilter\_v3) into list, this allows mergeing

multiple dataset this way.

labels a vector of labels for the samples.

thr One of the following "Total", "VerySensitive", "Sensitive", "Specific"

qualifiedCellCut

The minimum median mitochondrial coverage for a qualified cell, default is 10

OnlyHetero If only consider the heteroplasmy variants, default is T

VAFcut only use variants with VAF smaller than VAFcut. Default is 1. We can use

smaller value to constrain into only using rare variants

Cellcut only use variants with at least cellcut cells carry

maxctscut only use variants with at least in one cell with at leaset maxctscut variant frag-

ments

#### Value

redeemR class

Create\_redeemR

Create\_redeemR

### **Description**

This function is to create redeemR with basic information

# Usage

```
Create_redeemR(
   VariantsGTSummary = VariantsGTSummary,
   qualifiedCellCut = 10,
   VAFcut = 1,
   Cellcut = 2,
   maxctscut = 2
)
```

### Arguments

```
VariantsGTSummary
```

simply put GTSummary (Generated by redeemR.read)

qualifiedCellCut

The minimum median mitochondrial coverage for a qualified cell, default is 10

CV 15

VAFcut only use variants with VAF smaller than VAFcut. Default is 1. We can use

smaller value to constrain into only using rare variants

Cellcut only use variants with at least cellcut cells carry

maxctscut only use variants with at least in one cell with at leaset maxctscut variant frag-

ments

 ${\tt Only Hetero} \qquad \text{If only consider the heteroplasmy variants, default is } T$ 

#### Value

redeemR class

CV Internal CV

# Description

This function allows you to read raw data from XX/final folder, the output from mitoV

# Usage

CV(x)

# Arguments

x input a vector of numeric values

Datatoplots-class An intermediate S4 class Datatoplots

# Description

An intermediate S4 class Datatoplots

### **Slots**

clustering dataframe that store the data to plot

16 DepthSummary

DE.gettripple	DE.gettripple	
---------------	---------------	--

# Description

This function is to prepare the data format that is used to differentially expression calling. It include the raw matrix; data.info and size effect

### Usage

```
DE.gettripple(datapair, cpcol, withscran = F)
```

#### **Arguments**

datapair tyhe datapair generated from datapair.mk cpcol The column name for comparison.

withscran if true, use deconvolution to calculate size effect.

#### Value

This will return .tri.dummy file that is the input for DE analysis

### **Examples**

```
ROCKvsnorock.endo.tri.dummy<-DE.gettripple(ROCKvsnorock.endo.paired,cpcol="name")
```

DepthSummary	Function to summarize the depth (Total that passed Q30)

# Description

This function allows you to summarize the depth

#### Usage

```
DepthSummary(path, CellSubset = NA, only_Total = T)
```

The XX/final folder, the output from mitoV

# **Arguments**

path

Processed

paen	The 122 mai forces, the output from the v
CellSubset	A vector of ATAC cell names for subsetting, default is NA
only_Total	Default is T, Only return total depth summary. Don't care about depth in different quality filtering

Boolean variable(Default T), if true directly readRDS("depth.RDS") or, generate

and saveout "depth.RDS"

### Value

this returns depth which is a list of 4 list(Total/VerySensitive/Sensitive/Specific), each contains 2 df, summarize mito coverage by Pos/Cell

df2ProfileMtx 17

#### **Examples**

WD<-"/lab/solexa\_weissman/cweng/Projects/MitoTracing\_Velocity/SecondaryAnalysis/Donor01\_CDN1CD34\_1.depth<-DepthSummary(WD,Processed = T)

df2ProfileMtx

This is a convinience function, internal

## **Description**

This is a convinience function, internal

# Usage

```
df2ProfileMtx(df)
```

```
DistObjects-class An intermediate S4 class Datatoplots
```

### Description

An intermediate S4 class Datatoplots

#### **Slots**

```
jaccard distance object dist: Jaccard distance
Dice distance object dist: Dice distance
jaccard3W distance object dist: jaccard3W
```

DoDE

DoDE

# Description

This is the main function for calculating differentially expressed genes

#### Usage

```
DoDE(tri.dummy, cpcol, onlyoneSample = F, cpus = 16)
```

### **Arguments**

tri.dummy this is generated from DE.gettripple

cpcol the column in tri.dummy\$info, the contents of which are used for iteratively

compare with one another

onlyoneSample

If true, regress out batch effect. Notice, there should be a "Sample" column in

in tri.dummy\$info that indicate sample or donor or batch

cpus a number of cpus being used for calculation, default is 16

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#### Value

return a list that includes all DE result iteratively

#### **Examples**

ROCKvsnorock.endo.de<-DoDE (ROCKvsnorock.endo.tri.dummy, "name", onlyoneSample=T, cpus=16)

FromDist2Graph

From Dist2Graph From distance object or matrix to graph, default is to return igraph object This function was developed based on

### Description

FromDist2Graph From distance object or matrix to graph, default is to return igraph object This function was developed based on

### Usage

```
FromDist2Graph(d, k.param = 30, return_igraph = T)
```

#### **Arguments**

d the distance matrix, this can be either dist or a matrix

k.param K default is 30

return\_igraph

Wheather return igraph, default is T which return igraph. Otherwise, return adjacent matrix

#### Value

igraph or adjacent matrix

GEM\_Wrapper

Wrap Seurat RNA clustering

# Description

This function allows you to perform standard sc-RNA clustering

# Usage

```
GEM_Wrapper(mtx = bmmc.data$`Gene Expression`, exp = "DN1_BMMC1", res = 0.5)
```

# Arguments

mtx sparse Matrix of class "dgCMatrix", each row is a gene, each column is a cell,

res The name of this sample/experiment clustering resolution, default=0.5

get\_ancestral\_nodes 19

#### Value

this returns seurat object with RNA clustering

### **Examples**

```
bmmc.data=Read10X(data.dir = "XX/CellRanger/Donor01_BMMC_1/outs/filtered_feature_bc_matri
docluster_GEM(mtx=bmmc.data$`Gene Expression`,exp="DN1_BMMC1")
```

```
get_ancestral_nodes
```

This is a convinience function, internal borrowed from https://github.com/NickWilliamsSanger/treemut/blob/main/R/treemut.R#L68

### **Description**

This is a convinience function, internal borrowed from https://github.com/NickWilliamsSanger/treemut/blob/main/R/tree

### Usage

```
get_ancestral_nodes(node, edge, exclude_root = TRUE)
```

```
Get_Clonal_Variants
```

Get\_Clonal\_Variants

### **Description**

This function identify specific mutations for each clone based on Fisher Exact Test Of note, the ReDeeM object need to have Clone\_merge in CellMeta (After running Add\_tree\_cut)

### Usage

```
Get_Clonal_Variants(object)
```

# Arguments

object ReDeeM object

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GTSummary

Function to generate GTS summary

# Description

This function allows you to summarize the meta data for each genotyped variant

### Usage

```
GTSummary (RawGenotypes, filterN = T)
```

#### **Arguments**

```
RawGenotypes Well-named "RawGenotypes.Sensitive.StrandBalance" file in function CW_mgatk.read filterN Boolean variable, if true filter out the variant with "N"
```

#### Value

Genotypes.summary a dataframe that summarize several metrics for each genotype

# **Examples**

```
Usually used inside of function CW_mgatk.read
```

LineageBiasPlot

plot\_npSummary to plot the lineage composition

# Description

plot\_npSummary to plot the lineage composition

# Usage

```
LineageBiasPlot(npresult, pre)
```

### **Arguments**

npresult from ProgenyMapping\_np

pre Any short description for this plot to print with the plot

MakeAllNodes 21

MakeAllNodes	Define a function make the Allnodes(Node\Parent\Freq\CladeSize),
	where Freq is the number of variants assigned to the node(as ending
	point) from redeemR object,

#### **Description**

Define a function make the Allnodes(NodelParentlFreqlCladeSize), where Freq is the number of variants assigned to the node(as ending point) from redeemR object,

#### Usage

```
MakeAllNodes(redeemR = DN4_stemcell_redeemR.seed.verysensitive, prob.cut = 0.3)
```

#### **Arguments**

redeemR a redeemR object already have the tree built

prob.cut The probability cutoff to include confidently assigned variant

MakeDF4Regress Define a function to make two dataframe for regression analysis This function was developed based on HSC\_multiome\_Het\_2.ipynb

### **Description**

MakeDF4Regress Define a function to make two dataframe for regression analysis This function was developed based on HSC\_multiome\_Het\_2.ipynb

### Usage

```
MakeDF4Regress(
   multiome_wrapper = Donor04_HSC_Multiome_wrapper,
   redeemR = DN4_stemcell_redeemR.seed.sensitive,
   progeny_np = DN4_HSC_LSI_progeny_np,
   assay = "SCT",
   useNPimputation = T,
   maxcloneUMI = 10
)
```

#### **Arguments**

```
multiome_wrapper
```

This outject should includes all and more than HSCs cells in redeemR

redeemR scredeemR object for HSC
progeny\_np run via ProgenyMapping\_np
assay SCT for expression, ATAC for ATAC

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useNPimputation

default is T, use all cells called by network propagation, inaddition to the top cells in redeemR

maxcloneUMI

default is 10, Only include genes, in the max clone the expression greater than

#### Value

list(mtx.clone=mtx.clone,mtx.clone.norm.scale=mtx.clone.norm.scale)

Define a function to make nn list, which can be further used to make adjacency matrix This scan row by row, looking for k.param nearest neighbours

# Description

Define a function to make nn list, which can be further used to make adjacency matrix This scan row by row, looking for k.param nearest neighbours

#### Usage

```
MakeNN(d, k.param = 15)
```

# Arguments

d Distance matrix, can be a dist object or matrix

k.param Default is 15

# Value

return an nn list, which has two components: nn\$idx and nn\$dist

Make\_AnnTable

Make\_AnnTable, Make a big dataframe, each row is a cell, each column includes info such as clonal UMAP, Clonal ID, ATAC/RNA/WNN UMAP, PCA, gene expression of chosen gene, etc. Require a redeemR object and a multiome wrapper that better matches the cells in the redeemR

# Description

Make\_AnnTable, Make a big dataframe, each row is a cell, each column includes info such as clonal UMAP, Clonal ID, ATAC/RNA/WNN UMAP, PCA, gene expression of chosen gene, etc. Require a redeemR object and a multiome wrapper that better matches the cells in the redeemR

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#### Usage

```
Make_AnnTable(
  redeemR = DN4_HSC_redeemR.Sensitive,
  Multiome = Donor04 HSC Multiome wrapper,
  clonal_features = c("nCount_mitoV", "seurat_clusters"),
  clonal_features_rename = c("nCount_mitoV", "clone_clusters"),
  CellMeta_features = c("meanCov", "nCount_RNA", "nFeature_RNA", "nCount_ATAC",
    "nFeature_ATAC", "CellType"),
  CellMeta_features_rename = c("Mito_meanCov", "nCount_RNA", "nFeature_RNA",
    "nCount_ATAC", "nFeature_ATAC", "CellType"),
  multiome_features = c("seurat_clusters"),
  multiome_features_rename = c("NewSeurat_cluster"),
  RNAUMAP = T,
  ATACUMAP = T
  WNNUMAP = T
  PCA = F,
  LSI = F,
  Variants = "",
  genes = "",
  peaks = "",
  PostTrans_from = c(2, 3),
  PostTrans_to = c(2, 1)
```

#### **Arguments**

LSI

```
eg. DN4_HSC_redeemR.Sensitive
redeemR
                eg. Donor04_HSC_Multiome_wrapper, Multiome_wrapper object that matches
Multiome
                with the redeemR, a reclustering using Multi_Wrapper() is recommended
clonal_features
                eg. c("nCount mitoV", "seurat clusters"), The column names take from redeemR@Seurat@meta.data
                importantly the clonal clusterings
clonal_features_rename
                eg. c("nCount_mitoV","clone_clusters") Rename the clonal_features
CellMeta_features
                eg. c("meanCov","nCount_RNA","nFeature_RNA","nCount_ATAC","nFeature_ATAC","CellType")
                The column names take from redeemR@CellMeta, may useful cell features
CellMeta_features_rename
                eg. c("Mito_meanCov","nCount_RNA","nFeature_RNA","nCount_ATAC","nFeature_ATAC","Cell'
                Rename the CellMeta
multiome_features
                eg. c("seurat_clusters") The column names take from Multiome@meta.data
multiome_features_rename
                eg. c("NewSeurat_cluster") Rename the column names for multiome_features
                default T
RNAUMAP
                Default T
ATACUMAP
WNNUMAP
                Default T
PCA
                Default T
```

Default T

24 Make\_Cells4Nodes

```
Variants Default "" can be a vector of variant names format is eg "Variants10020TC"

genes Default "" can be a vector of gene names, for example c("HLF","CD34")

peaks Default "" can be a vector of peaks names

PostTrans_from

Default c(2,3) # This is a tricky part eh nmerging files are involved, find the postfix from cellranger agg for different sample

PostTrans_to Default c(2,1)
```

#### Value

AnnTable

### **Description**

Define a function to make a list, each contains the cell names for a node

#### Usage

```
Make_Cells4Nodes(
   tr = DN4_SLCT_HSC_w_jaccard.njtree@phylo,
   min.node.size = 10,
   max.node.fra = 0.33
)
```

# Arguments

```
tr phylo object (ape)

min.node.size

default is 10, only the nodes with more than 10 tips are included (# Minimum # tips in the node to be included)

max.node.fra

default is 0.33, only consider the nodes with less than max.node.fra*total cell number (# The up limit of the node size(Fraction of all tips) to be considered)
```

### Value

return a list each contains the cell names for a node that meets the criteria

Make\_matrix 25

Make\_matrix This will make the matixies of Cell VS mitochondrial variants and return redeemR Results stored in Cts.Mtx and Cts.Mtx.bi

### **Description**

Make\_matrix This will make the matixies of Cell VS mitochondrial variants and return redeemR Results stored in Cts.Mtx and Cts.Mtx.bi

#### Usage

```
Make_matrix(object, ...)
```

#### **Arguments**

object redeemR class

Make\_matrix, redeemR-method

Make\_matrix This will make the matixies of Cell VS mitochondrial variants and return redeemR Results stored in Cts.Mtx and Cts.Mtx.bi

### **Description**

Make\_matrix This will make the matixies of Cell VS mitochondrial variants and return redeemR Results stored in Cts.Mtx and Cts.Mtx.bi

# Usage

```
## S4 method for signature 'redeemR'
Make_matrix(object, onlyhetero = T)
```

# Arguments

object redeemR class

onlyhetero Only use heteroplasmic mutations

#### Value

redeemR class

Make\_tree This will generate a basic phylogenetic tree

### **Description**

Make\_tree This will generate a basic phylogenetic tree

### Usage

```
Make_tree(object, d = "jaccard", algorithm = "upgma", onlyreturntree = F, ...)
```

# **Arguments**

object redeemR class

d "jaccard" or "Dice" or "jaccard3W"

algorithm the algorithm used to build the tree, choose from "nj" and "upgma"

Make\_tree, redeemR-method

Make\_tree This will generate a basic phylogenetic tree

# Description

Make\_tree This will generate a basic phylogenetic tree

# Usage

```
## S4 method for signature 'redeemR'
Make_tree(object, d, algorithm, onlyreturntree = F)
```

### **Arguments**

object mitoTracin class

d "jaccard" or "Dice" or "jaccard3W" or "w\_jaccard" "w\_cosine" "LSIdist"

algorithm the algorithm used to build the tree, choose from "nj" and "upgma"

#### Value

mitoTracin class

MergeMtx 27

MergeMtx

Function to Merge sparse Matrix

#### **Description**

This function allows you to input a list of sparse matrix and merge by rownames, return a new sparse matrix

### Usage

```
MergeMtx(mtx.list, postfix)
```

### **Arguments**

mtx.list A list of sparse matrix to be merged

postfix a vector of postfix (Usually are numbers that added at the end of cell names).

Better be consistent with a merged redeemR object orders

#### Value

new sparse matrix

#### **Examples**

```
Donor4_HSC_HPC_BMMC.Mtx<-MergeMtx(list(Donor04_BMMC_Multiome_wrapper$seurat@assays$RNA@ccDonor4_HSC_HPC_BMMC.RNA.seurat<-GEM_Wrapper(Donor4_HSC_HPC_BMMC.Mtx)
```

Motifenrich.binom

Motifenrich.binom In house function to compute enrichment from Fimo This function was developed based on HSC\_multiome\_Het.ipynb and HSC\_multiome\_Het\_2.ipynb

# **Description**

Motifenrich.binom In house function to compute enrichment from Fimo This function was developed based on HSC\_multiome\_Het.ipynb and HSC\_multiome\_Het\_2.ipynb

#### Usage

```
Motifenrich.binom(queryP.motif, controlP.motif, alt = "greater")
```

# Arguments

28 MutationProfile.bulk

Multi\_Wrapper

Wrap Seurat Multiomics clustering

### **Description**

This function allows you to perform standard sc-multiome clustering

#### Usage

```
Multi_Wrapper(
    path = "/lab/solexa_weissman/cweng/Projects/MitoTracing_Velocity/SecondaryAr
    atacmin = 1000,
    umimin = 1000,
    CellID = NULL
)
```

#### **Arguments**

path this should be the path to the cell-ranger results XX/outs

atacmin minimum atac fragment for each cell, default is 1000

umimin minimum rna umi for each cell, default is 1000

cellID to be used for input(useful for re-clustering), default is NULL which will use the info from path/per\_barcode\_metrics.csv

#### Value

this returns seurat object with both RNA and ATAC

# **Examples**

```
Multi_Wrapper(path="XX/CellRanger/Donor01_BMMC_1/outs/")
```

MutationProfile.bulk

Function to plot bulk level mutation signatures

### **Description**

This function allows you to plot the mito mutation signatures

### Usage

```
MutationProfile.bulk(cell_variants)
```

### **Arguments**

```
cell_variants
```

a vector of variants formated as c('93\_A\_G"103\_G\_A"146\_T\_C'

NN2M 29

#### Value

```
p from ggplot2
```

#### **Examples**

```
MutationProfile.bulk(DN1CD34_1.Variants.feature.lst[[name]]$Variants
```

NN2M

Define a function convert nn list to adjacency matrix that can be further used for igraph

#### **Description**

Define a function convert nn list to adjacency matrix that can be further used for igraph

### Usage

```
NN2M(nn)
```

### **Arguments**

nn

nn list, which has two components: nn\$idx and nn\$dist

#### Value

return an nn.matrix. This is adjacency matrix can be input to igraph graph<-graph\_from\_adjacency\_matrix(nn.matrix,dia = F,mode = "undirected")

plot\_depth

Function to plot the mito depth summary

### **Description**

This function allows you to plot both position-wise and cell-wise mito depth summary

### Usage

```
plot_depth(ob, name = "", w = 10, h = 3)
```

#### **Arguments**

ob The redeemR object

name The plot name shown on top
w the Width of the plot, default=10
h the height of the plot default=3

#### Value

directly out put the plot

30 plot\_variant

#### **Examples**

```
plot_depth(DN1CD34_1.depth$Total,"Total")
```

plot\_npSummary

plot\_npSummary to assess the outputlevel

#### **Description**

plot\_npSummary to assess the outputlevel

#### Usage

```
plot_npSummary(npresult, orderby = "Total.norm", pre)
```

# **Arguments**

npresult from ProgenyMapping\_np

orderby Normalize by, so far can work with "Total.norm" and "Total.norm\_NPadj"

pre Any short description for this plot to print with the plot

plot\_variant Function to plot variant metrics

# Description

This function allows you to plot the mito mutation metrics For each category(stringency), p1: Variant allele frequency(VAF); p2: Heteroplasmy histogram p3: CellN(Number of caells that carry the variants) VS maxcts( The number of variant counts in the highest cell) p4: Histogram to show the distribution of the number of variant per cell

### Usage

```
plot_variant(ob, p4xlim = 50, QualifyCellCut = 10)
```

# **Arguments**

ob The redeemR object  $p4xlim \qquad \text{the p4 xlim(number of variant per cell), default is 50} \\ \text{QualifyCellCut}$ 

median coverage for qualified cells, default is 10

#### Value

no returns, directly plot

# **Examples**

```
plot_variant(DN1CD34_1.VariantsGTSummary,DN1CD34_1.Variants.feature.lst,depth=DN1CD34_1.c
```

ProgenyMapping 31

ProgenyMapping

Define a function to perform single-cell based hard porogeny assignment This function was developed based on DN4T2.basics.ipynb

### **Description**

Define a function to perform single-cell based hard porogeny assignment This function was developed based on DN4T2.basics.ipynb

### Usage

```
ProgenyMapping(
   HSC_redeemR = DN4_PhenoHSC_redeemR.verysensitive,
   Full_redeemR = DN4_BMMC_HSPC_HSC_redeemR.verysensitive,
   distCut = 0.95,
   d = "w_jaccard"
)
```

#### **Arguments**

HSC\_redeemR The HSC\_redeemR is the redeemR object for defined HSC

Full\_redeemR The FULL\_redeemR is the redeemR object for the full BMMC\_HSPC\_HSC

distCut Default is 0.95, the distance, below which I define as the related progeny

ProgenyMapping\_np

ProgenyMapping\_np Define a function to compute network propagation based probability FromDist2Graph is needed to convert fistance matrix into MNN graph

### Description

ProgenyMapping\_np Define a function to compute network propagation based probability FromDist2Graph is needed to convert fistance matrix into MNN graph

#### Usage

```
ProgenyMapping_np(
   HSC_redeemR = DN4_stemcell_redeemR.seed.verysensitive,
   Full_redeemR = DN4_BMMC_HSPC_HSC_redeemR.verysensitive,
   CloneCol = "Clone_merge",
   k = 30,
   gm = 0.5,
   useLSI = F,
   useSCAVENGE_LSI = F,
   subsample = F,
   ProbCut = 0.7,
   Celltype = "Rig.CellType"
)
```

32 quick\_w\_cosine

### **Arguments**

HSC\_redeemR The HSC\_redeemR is the redeemR object for defined HSC, have already gone

through Add\_DepthMatrix-Add\_AssignVariant-Add\_tree\_cut, otherwise, need

othereise, need a column in CellMeta that indicates the clone ID

Full\_redeemR The FULL\_redeemR is the redeemR object for the full BMMC\_HSPC\_HSC

CloneCol "Clone\_merge"

k the k.param used for MNN graph

gamma default is 0.05 which mean 95% information is passing out

ProbCut The cutoff of the maximum probability for a given progeny cell(If the maximum

probability is lower than ProbCut, it will be filtered)

Celltype The column to be used in aggregate into lineages

#### Value

a list of two ALLmeta.npClone (A meta data with last column npClone), np\_mat (the network propagation matrix))

quick\_w\_cosine

Compute weighted cosine distance

### **Description**

Compute weighted cosine distance

#### Usage

```
quick_w_cosine(M, w)
```

#### **Arguments**

M the binary matrix, Each row is a cell, each column is a variant, generated by

 $Make\_matrix$ 

w weight for each variant, a vector

#### Value

dist object

quick\_w\_jaccard 33

quick\_w\_jaccard

Compute weighted jaccard distance

# Description

Compute weighted jaccard distance

# Usage

```
quick_w_jaccard(M, w)
```

# **Arguments**

M the binary matrix, Each row is a cell, each column is a variant, generated by

Make\_matrix

w weight for each variant, a vector

# Value

dist object

Reclustering

Function to reclustering a seurat object

# Description

This function allows you to input a seurat object(multiome), redo clustering. Usually this is after subset

# Usage

```
Reclustering(ob)
```

# Arguments

ob

a seurat object

#### Value

a seurat object

Reclustering\_hm

Function to reclustering\_hm a seurat object with Harmony

### **Description**

This function allows you to input a seurat object(multiome), redo clustering harmony by a certain column in meta data. Usually this is after subset

### Usage

```
Reclustering_hm(
  ob = DN4_RigHSC_T1T2_Multiome_wrapper_filtered.anno,
  HarmonyBy = "TimePoint"
)
```

### **Arguments**

ob a seurat object

HarmonyBy The columne name in meta that will be used for Harmony

#### Value

a seurat object

```
reconstruct_genotype_summary
```

This is a function borrowed from https://github.com/NickWilliamsSanger/treemut/blob/main/R/treemut.R#L68 Input phylo object, return a "profile matrix"—Edge(or denoted as the ending node) vs cell. a 0, 1 character string that indicate what cells in a given node

#### **Description**

This is a function borrowed from https://github.com/NickWilliamsSanger/treemut/blob/main/R/treemut.R#L68 Input phylo object, return a "profile matrix"–Edge(or denoted as the ending node) vs cell. a 0, 1 character string that indicate what cells in a given node

#### Usage

```
reconstruct_genotype_summary(phylo)
```

#### **Arguments**

phylo phylo an ape object

#### Value

df includes df\$df which is a big data frame, and df\$sample that is the cell names

redeemR-class 35

redeemn-crass major redeem class that store clothal-resolved manti-omics	redeemR-class	Major redeem class that store clonal-resolved multi-omics	
--	---------------	---	--

#### **Description**

Major redeem class that store clonal-resolved multi-omics

#### **Slots**

GTsummary.filtered The Mitochondrial genotype data frame

CellMeta Store meta data for each cell type

V.fitered.list a list of data frame of variant metrics, VAF, cellN, etc (each for different stringency),

UniqueV A character showing the number of usable variant

Cts.Mtx A sparse matrix cell-mitoVariants, store the variant count

 $\label{thm:count} \text{Cts.Mtx.bi} \ \ A \ sparse \ matrix \ cell-mitoVariants, \ The \ variant \ count \ has \ been \ binarized \ into \ 0 \ and \ 1$ 

Ctx.Mtx.depth A sparse matrix cell-mitoVariants(total counts for each position), store the variant count

para A character showing the parameter of this object

Seurat Seurat object storing the clonal clustering results

DataToplotList The customized class of Datatoplots: A list of dataframe for further plotting

DistObjects The customized class that stores the cell-cell distances

TREE The customized class that wraps phylogenetic tree

redeemR.read Function to read in mitoV outputs

#### **Description**

This function allows you to read raw data from XX/final folder, the output from mitoV

#### Usage

```
redeemR.read(path, thr = "S", Processed = F)
```

# **Arguments**

path The XX/final folder, the output from mitoV

thr The thredhold of filtering T(Total),LS(Less Stringent:c=0.75,a1=2,a2=1), S(Stringent:c=0.75,a1=3,a2=1),

VS(Very Stringent:c=0.75,a1=4,a2=3)"

Processed Boolean variable (Default F), if true directly readRDS("VariantsGTSummary.RDS")

or, generate and saveout "VariantsGTSummary.RDS"

36 Runplot\_scale\_3

#### Value

this returns depth which is a list of 4 df (Total/VerySensitive/Sensitive/Specific), each is a genotype summary

### **Examples**

```
WD<-"/lab/solexa_weissman/cweng/Projects/MitoTracing_Velocity/SecondaryAnalysis/Donor01_CDN1CD34_1.VariantsGTSummary<-CW_mgatk.read(WD,Processed =T)
```

Runplot\_scale\_2

plot\_npSummary to assess the outputlevel vs lineage bias, normalize by assigned

#### **Description**

plot\_npSummary to assess the outputlevel vs lineage bias, normalize by assigned

#### Usage

```
Runplot_scale_2(
   datatoplot = DN4_HSC_LSI_progeny$LineageSummary$output_lineage.summary.pct.sca
   pre
)
```

#### **Arguments**

datatoplot A slot from the result of ProgenyMapping\_np: datatoplot.scale pre Any short description for this plot to print with the plot

Runplot\_scale\_3

plot\_npSummary to assess the outputlevel vs lineage bias, normalize by HSC original clone size

#### **Description**

plot\_npSummary to assess the outputlevel vs lineage bias, normalize by HSC original clone size

### Usage

```
Runplot_scale_3(
   datatoplot = DN4_HSC_LSI_progeny$LineageSummary$output_lineage.summary.pct.sca
   pre
)
```

## **Arguments**

datatoplot A slot from the result of ProgenyMapping\_np: datatoplot.scale pre Any short description for this plot to print with the plot

Run\_Lin\_regression 37

```
Run_Lin_regression Run_Lin_regression
```

### **Description**

Firstly used in HSC\_multiome\_Het\_2.ipynb

## Usage

```
Run_Lin_regression(
   LinOut,
   regress_factor = c("OutLevel.scale", "OutLevel_NPadj.scale", "Lym", "Mye", "MK"),
   n.cores = 8
)
```

### **Arguments**

```
\label{linOut_produced_by MakeDF4Regress} $$ regress_factor $$ default is c("OutLevel.scale","OutLevel_NPadj.scale","Lym","Mye","MK","ME") $$ n.cores $$ default is 8$
```

```
Run_Lin_regression_poi
```

Run\_Lin\_regression\_poi Firstly used in HSC\_multiome\_Het\_2.ipynb This function was developed based on

### Description

Run\_Lin\_regression\_poi Firstly used in HSC\_multiome\_Het\_2.ipynb This function was developed based on

# Usage

```
Run_Lin_regression_poi(
   LinOut,
   regress_factor = c("OutLevel.scale", "OutLevel_NPadj.scale", "Lym", "Mye", "MK"),
   n.cores = 8
)
```

### **Arguments**

```
LinOut produced by MakeDF4Regress
regress_factor
default is c("OutLevel.scale","OutLevel_NPadj.scale","Lym","Mye","MK","ME")
n.cores =8
```

SeuratLSIClustering

SeuratLSIClustering This will use the mito variants for Seurat clustering (LSI based)

### **Description**

SeuratLSIClustering This will use the mito variants for Seurat clustering (LSI based)

### Usage

```
SeuratLSIClustering(object, ...)
```

### **Arguments**

object redeemR class

SeuratLSIClustering, redeemR-method

SeuratLSIClustering This will use the mito variants for Seurat clustering (LSI based)

# Description

SeuratLSIClustering This will use the mito variants for Seurat clustering (LSI based)

# Usage

```
## S4 method for signature 'redeemR'
SeuratLSIClustering(
  object,
  binary = T,
  res = 0.6,
  lsidim = 2:50,
  rmvariants = c("Variants310TC", "Variants3109TC", "Variants5764CT"))
```

# **Arguments**

binary

Default is tree, to make use of the binary matrix

res

Default os 0.3, the resolution of the clustering

redeemR

class

### Value

redeemR class

show,redeemR-method 39

```
show, redeemR-method
```

show This will show the basics of redeemR class

### **Description**

show This will show the basics of redeemR class

### Usage

```
## S4 method for signature 'redeemR'
show(object)
```

### **Arguments**

object

redeemR class

#### Value

print out basics

Show\_Consensus

Function to plot consensus mtDNA mutation benchmark

### **Description**

This function allows you to plot the mito mutation consensus levels It will print out Quantiles of UMI family size; Quantile of consensus score; Percentage of R1/R2 overlaped mutation detections It will also plot random N mutations as examples to show consensus metrics

# Usage

```
Show_Consensus(ob, N = 25)
```

### **Arguments**

ob	The redeemR object
N	number of example variants to show, default is 25

split\_profile

This is a convinience function, internal borrowed from https://github.com/NickWilliamsSanger/treemut/blob/main/R/treemut.R#L68

# Description

This is a convinience function, internal borrowed from https://github.com/NickWilliamsSanger/treemut/blob/main/R/tree

# Usage

```
split_profile(profile)
```

40 Subset\_redeemR

	_		
st	r277e	ct	or

This is a convinience function, internal

# Description

This is a convinience function, internal

### Usage

```
str2vector(x)
```

Subset\_redeemR

Subset\_redeemR Subset a redeemR object by selecting a subset of cells, return a new redeemR object with only 4 slots: para; CellMeta; Cts.Mtx.bi; UniqueV, can be used for downstreme compute distance, clonal clustering, make tree, etc

### **Description**

Subset\_redeemR Subset a redeemR object by selecting a subset of cells, return a new redeemR object with only 4 slots: para; CellMeta; Cts.Mtx.bi; UniqueV, can be used for downstreme compute distance, clonal clustering, make tree, etc

## Usage

```
Subset_redeemR(redeemR, Cells, ExtraInfo = "Subset from ... ")
```

### **Arguments**

redeemR The Parent redeemR object eg. DN4\_HSC\_redeemR.Sensitive

Cells Important, give a vector of Cell names(ATAC cell names)

ExtraInfo Extra information, usually "Subset from ..."

# Value

redeemR Object

Tomerge\_v2 41

# Description

This function is to quickly merge two dataframe by rownames, but can choose to leave A or B all information

# Usage

```
Tomerge_v2(A, B, leavex = T, leavey = F)
```

### **Arguments**

A dataframe A

B dataframe B

# Value

return a data frame with merged information

# **Examples**

```
Tomerge_v2(A,B)
```

 ${\tt Translate\_RNA2ATAC} \ \ \textit{Function to translate the RNA barcode into ATAC barcode and add a } \\ column$ 

# Description

This function allows you to input the metadata with row name as cell barcode

# Usage

```
Translate_RNA2ATAC(
  meta = bmmc.filtered@meta.data,
  PostFix = T,
  bclength = 16,
  from = c(1, 2, 3),
  to = c(1, 2, 3))
```

#### **Arguments**

meta a dataframe with the row names as the RNA cell barcode usually with the post

-1

bclength The cell barcode length, default is 16

from A vector of the postfix, usually is c(1,2,3,...), it depends on how many samples

are aggregated in Cellranger RNA part

A vector of the postfix, those cooresponds to the postfix added in redeemR,

in general, if it matches, then simply c(1,2,3,...), but in case not match, here

provides a way to transform into scredeemR order

#### Value

meta a dataframe

#### **Examples**

```
Translate_RNA2ATAC(meta)
```

```
Translate_simple_ATAC2RNA
```

Translate\_simple\_ATAC2RNA

#### **Description**

This function allows you to input the ATAC name to translate to RNA name

# Usage

```
Translate_simple_ATAC2RNA(
  name,
  PostFix = T,
  bclength = 16,
  from = c(1, 2, 3),
  to = c(1, 2, 3))
```

#### **Arguments**

name RNA name, as the RNA cell barcode usually with the post -1

bclength The cell barcode length, default is 16

from A vector of the postfix, usually is c(1,2,3,...), it depends on how many samples

are aggregated in Cellranger RNA part

A vector of the postfix, those cooresponds to the postfix added in redeemR,

in general, if it matches, then simply c(1,2,3,...), but in case not match, here

provides a way to transform into redeemR order

#### Value

RNA name Translate\_RNA2ATAC(a vector of RNA names)

# Description

This function allows you to input the RNA name to translate to ATAC name

# Usage

```
Translate_simple_RNA2ATAC(
  name,
  PostFix = T,
  bclength = 16,
  from = c(1, 2, 3),
  to = c(1, 2, 3))
```

### **Arguments**

name	RNA name, as the RNA cell barcode usually with the post -1
bclength	The cell barcode length, default is 16
from	A vector of the postfix, usually is $c(1,2,3,)$ , it depends on how many samples are aggregated in Cellranger RNA part
to	A vector of the postfix, those cooresponds to the postfix added in redeemR, in general, if it matches, then simply $c(1,2,3,)$ , but in case not match, here provides a way to transform into redeemR order

### Value

ATAC name Translate\_RNA2ATAC(a vector of RNA names)

TREE-class

An intermediate S4 class Tree that store tree info

# Description

An intermediate S4 class Tree that store tree info

### **Slots**

```
phylo the phylo tree class from ape package
treedata treedata class from tidytree
records character to store annotations
```

Vfilter\_v4

Vfilter\_v3

Function to filter variants, deprecated

### **Description**

This function allows you to filter variants, deprecated, use Vfilter\_v4 instead

# Usage

```
Vfilter_v3(
   InputSummary,
   depth,
   Rmvhomo = F,
   Min_Cells = 2,
   Max_Count_perCell = 2,
   QualifyCellCut = 10
)
```

### Arguments

InputSummary The GTSummary file read in by function CW\_mgatk.read

depth The .depth file by function DepthSummary

Rmvhomo Boolean (Default F) If true, remove the homozygous variants

Min\_Cells Default 2, A qualified variant needs the minimum number of cells that have this

variant

Max\_Count\_perCell

Default 2, A qualified variant needs to show at least 2 counts in one cell

QualifyCellCut

Default 10, Minimum depth for a qualified cell

#### Value

this returns feature.list

# **Examples**

```
DN1CD34_1.Variants.feature.lst<-Vfilter_v3(InputSummary=DN1CD34_1.VariantsGTSummary,depth
```

Vfilter\_v4

Function to filter variants, v4

# Description

This function allows you to filter variants, deprecated, use Vfilter\_v4 instead

Vfilter\_v4 45

### Usage

```
Vfilter_v4(
    InputSummary = VariantsGTSummary,
    Min_Cells = 2,
    Max_Count_perCell = 2,
    QualifyCellCut = 10
)
```

# Arguments

InputSummary The GTSummary file read in by function CW\_mgatk.read

Min\_Cells Default 2, A qualified variant needs the minimum number of cells that have this

variant

Max\_Count\_perCell

Default 2, A qualified variant needs to show at least 2 counts in one cell

QualifyCellCut

Default 10, Minimum depth for a qualified cell

depth The .depth file by function DepthSummary

Rmvhomo Boolean (Default F) If true, remove the homozygous variants

# Value

this returns feature.list

### **Examples**

DN1CD34\_1.Variants.feature.lst<-Vfilter\_v3(InputSummary=DN1CD34\_1.VariantsGTSummary,depth

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