

# Package ‘uSORT’

September 16, 2024

**Title** uSORT: A self-refining ordering pipeline for gene selection

**Version** 1.30.0

**Author** Mai Chan Lau, Hao Chen, Jinmiao Chen

**Description** This package is designed to uncover the intrinsic cell progression path from single-cell RNA-seq data. It incorporates data pre-processing, preliminary PCA gene selection, preliminary cell ordering, feature selection, refined cell ordering, and post-analysis interpretation and visualization.

**Maintainer** Hao Chen <chen\_hao@immunol.a-star.edu.sg>

**biocViews** ImmunoOncology, RNASeq, GUI, CellBiology, DNASEq

**Depends** R (>= 3.3.0), tcltk

**VignetteBuilder** knitr

**Suggests** knitr, RUnit, testthat, ggplot2

**Imports** igraph, Matrix, RANN, RSpectra, VGAM, gplots, parallel, plyr, methods, cluster, Biobase, fpc, BiocGenerics, monocle, grDevices, graphics, stats, utils

**License** Artistic-2.0

**Encoding** UTF-8

**LazyData** true

**RoxygenNote** 5.0.1

**git\_url** <https://git.bioconductor.org/packages/uSORT>

**git\_branch** RELEASE\_3\_19

**git\_last\_commit** 9688ccc

**git\_last\_commit\_date** 2024-04-30

**Repository** Bioconductor 3.19

**Date/Publication** 2024-09-15

## Contents

autoSPIN . . . . .	3
clusterGenes1 . . . . .	4
compareModels1 . . . . .	5
differentialGeneTest1 . . . . .	5
diff_test_helper1 . . . . .	6
distance.function . . . . .	7
driving_force_gene_selection . . . . .	7
elbow_detection . . . . .	8
EXP_to_CellDataSet . . . . .	9
fluidigmSC_analyzeGeneDetection . . . . .	9
fluidigmSC_identifyExpOutliers . . . . .	10
fluidigmSC_isElementIgnoreCase . . . . .	11
fluidigmSC_readLinearExp . . . . .	12
fluidigmSC_removeGenesByLinearExpForAllType . . . . .	12
fluidigmSC_removeGenesByLinearExpForAllType_log2 . . . . .	13
monocle_wrapper . . . . .	13
neighborhood_sorting . . . . .	14
neighborhood_sortingcost . . . . .	15
neighborhood_sorting_wrapper . . . . .	15
pca_gene_selection . . . . .	16
Rwanderlust . . . . .	17
scattering_quantification_per_gene . . . . .	18
sorting_method_parameter_GUI . . . . .	18
SPIN . . . . .	19
STS_sorting . . . . .	20
STS_sortingcost . . . . .	20
STS_sorting_wrapper . . . . .	21
summed_local_variance . . . . .	22
summed_local_variance_cyclical . . . . .	22
summed_local_variance_linear . . . . .	23
sWanderlust . . . . .	23
trajectory_landmarks . . . . .	24
uSORT . . . . .	25
uSORT_GUI . . . . .	28
uSORT_parameters_GUI . . . . .	28
uSORT_preProcess . . . . .	29
uSORT_sorting_wrapper . . . . .	30
uSORT_write_results . . . . .	31
variability_per_gene . . . . .	32
wanderlust_wrapper . . . . .	32

## Index

34

---

autoSPIN	<i>A wrapper function for autoSPIN sorting method</i>
----------	---

---

### Description

A wrapper function for autoSPIN method which implements optimized local refinement using the selected SPIN sorting method, i.e. STS or Neighborhood.

### Usage

```
autoSPIN(data, data_type = c("linear", "cyclical"),
         sorting_method = c("STS", "neighborhood"), alpha = 0.2, sigma_width = 1,
         no_randomization = 20, window_perc_range = c(0.1, 0.9),
         window_size_incre_perct = 0.05)
```

### Arguments

data	An log2 transformed expression matrix containing n-rows of cells and m-cols of genes.
data_type	A character string indicating the type of progression, i.e. 'linear' (strictly linear) or 'cyclical' (cyclically linear).
sorting_method	A character string indicating the choice of SPIN sorting method, i.e. 'STS' (Side-to-Side) or 'Neighborhood'.
alpha	A fraction value denoting the size of locality used for calculating the summed local variance.
sigma_width	An integer number denoting the degree of spread of the gaussian distribution which is used for computing weight matrix for Neighborhood sorting method.
no_randomization	An integer number indicating the number of repeated sorting, each of which uses randomly selected initial cell position.
window_perc_range	A fraction value indicating the range of window size to be examined during local refinement.
window_size_incre_perct	A fraction value indicating the step size at each iteration for incrementing window size.

### Value

A data frame containing single column of ordered sample IDs.

**Examples**

```

set.seed(15)
da <- iris[sample(150, 150, replace = FALSE), ]
rownames(da) <- paste0('spl_',seq(1,nrow(da)))
d <- da[,1:4]
d1 <- da[,5,drop=FALSE]
res <- autoSPIN(data = d)
d1 <- d1[match(res$SampleID,rownames(d1)),]
annot <- data.frame(id = seq(1,nrow(res)), label=d1, stringsAsFactors = FALSE)
#ggplot(annot, aes(x=id, y=id, colour = label)) + geom_point() + theme_bw()

```

---

clusterGenes1	<i>A modified monocle's function</i>
---------------	--------------------------------------

---

**Description**

A modified monocle's function for 'compareModels' which identifies and removes genes whose reduced\_models is better than full\_models in term of likelihood

**Usage**

```

clusterGenes1(expr_matrix, krange, method = function(x) { as.dist((1 -
  cor(t(x)))/2) }, ...)

```

**Arguments**

expr_matrix	Expression matrix.
krange	krange.
method	method function.
...	Other parameters.

**Value**

test\_res a dataframe containing status of modeling and adjusted p-value

**Author(s)**

MaiChan Lau

---

compareModels1	<i>A modified monocle's function</i>
----------------	--------------------------------------

---

**Description**

A modified monocle's function for 'compareModels' which identifies and removes genes whose reduced\_models is better than full\_models in term of likelihood

**Usage**

```
compareModels1(full_models, reduced_models)
```

**Arguments**

full\_models a Monocle's vgam full model  
 reduced\_models a Monocle's vgam reduced/ null model

**Value**

test\_res a dataframe containing status of modeling and adjusted p-value

**Author(s)**

MaiChan Lau

---

differentialGeneTest1	<i>differential gene test</i>
-----------------------	-------------------------------

---

**Description**

modified from FludigmSC package

**Usage**

```
differentialGeneTest1(cds,
  fullModelFormulaStr = "expression~sm.ns(Pseudotime, df=3)",
  reducedModelFormulaStr = "expression~1", cores = 1)
```

**Arguments**

cds Input object.  
 fullModelFormulaStr Full model formula.  
 reducedModelFormulaStr Reduced model formula.  
 cores Number of cores will be used.

**Value**

test results

---

diff\_test\_helper1      *A modified monocle's helper function*

---

**Description**

A modified monocle's function for 'diff\_test\_helper1' which includes more attempts on finding models and also compute max. magnitude change in expression values predicted by GLM model

**Usage**

```
diff_test_helper1(x, fullModelFormulaStr, reducedModelFormulaStr,
  expressionFamily, lowerDetectionLimit = 0.1, type_ordering = "linear")
```

**Arguments**

x                      an expression data

fullModelFormulaStr      a Monocle's model structure

reducedModelFormulaStr    a Monocle's model structure

expressionFamily          a Monocle's family character

lowerDetectionLimit      a threshold value

type\_ordering          a character indicating the type of underlying cell progression, i.e. linear or circular

**Value**

test\_res a dataframe containing status of modeling and adjusted p-value

**Author(s)**

MaiChan Lau

---

distance.function	<i>A distance function A distance function computes cell-to-cell distance matrix.</i>
-------------------	---

---

**Description**

A distance function A distance function computes cell-to-cell distance matrix.

**Usage**

```
distance.function(expr, method = c("Euclidean", "Correlation", "eJaccard",  
    "none"))
```

**Arguments**

expr	An expression matrix containing n-rows of cells and m-cols of genes.
method	A character string indicating the distance function.

**Value**

A matrix containing n-by-n cell distance.

---

driving_force_gene_selection	<i>A feature/ gene selection function</i>
------------------------------	---

---

**Description**

A feature/ gene selection function (1) removes sparsely expressed genes, (2) identifies differentially expressed genes based on preliminary cell ordering, (3) removes highly dispersed genes from the identified DEGs, (4) further picks genes which are expected to have large expression difference on the 2 extreme ends of preliminary cell ordering

**Usage**

```
driving_force_gene_selection(cds, scattering.cutoff.prob = 0.75,  
    driving.force.cutoff = NULL, qual_cutoff = 0.05, min_expr = 0.1,  
    data_type = c("linear", "cyclical"), nCores = 1)
```

**Arguments**

**cds** a Monocle's CellDataSet object  
**scattering.cutoff.prob** probability used for removing largely dispersed genes  
**driving.force.cutoff** a value used for removing genes which do not change much along cell progress along cell progress path  
**qval.cutoff** a user-defined adjusted p-value below which genes are retained  
**min\_expr** the minimum expression value  
**data\_type** a character indicating the type of underlying cell progression, i.e. linear or cyclical.  
**nCores** Number of cores to use.

**Value**

integer

**Author(s)**

MaiChan Lau

**Examples**

```

dir <- system.file('extdata', package='uSORT')
file <- list.files(dir, pattern='.txt$', full=TRUE)
#exprs <- uSORT_preProcess(exprs_file = file)
#exp_raw <- t(exprs$exprs_raw)
#exp_trimmed <- t(exprs$exprs_log_trimmed)
#cds <- uSORT:::EXP_to_CellDataSet(exp_trimmed, exp_raw)
#driver_genes <- driving_force_gene_selection(cds = cds)

```

---

elbow\_detection      *A elbow detection function*

---

**Description**

A elbow detection function detects the elbow/knee of a given vector of values. Values will be sorted descendingly before detection, and the ID of those values above the elbow will be returned.

**Usage**

```
elbow_detection(scores, if_plot = FALSE)
```

**Arguments**

**scores** A vector of numeric scores.  
**if\_plot** Boolean determine if plot the results.



**Value**

a vector of selected elements IDs

**Examples**

```
scores <- c(10, 9, 8, 6, 3, 2, 1, 0.1)
elbow_detection(scores, if_plot = TRUE)
```

---

EXP\_to\_CellDataSet      *A function for constructing a Monocle's CellDataSet object from an expression matrix*

---

**Description**

A function for constructing a Monocle's CellDataSet object from an expression matrix

**Usage**

```
EXP_to_CellDataSet(log2_exp = NULL, expression_data_raw = NULL, lod = 1)
```

**Arguments**

log2\_exp              An log2 transformed expression matrix containing n-rows of cells and m-cols of genes.

expression\_data\_raw      A data frame containing raw expression values, with rownames of cells and colnames of genes.

lod                      A value of limit of detection in the unit of TPM/CPM/RPKM.

**Value**

A CellDataSet object.

---

fluidigmSC\_analyzeGeneDetection  
                                  *A gene detection function*

---

**Description**

A gene detection function computes the fraction of genes detected in each cell, reproduced from FluidigmSC package.

**Usage**

```
fluidigmSC_analyzeGeneDetection(expression_data, threshold = 1)
```

**Arguments**

expression_data	A data frame containing raw expression values, with rownames of genes and colnames of cells.
threshold	A limit of detection in the unit of TPM/CPM/RPMK.

**Value**

A data frame containing a column of number of genes detected, and a column of the corresponding percentage of gene detection, rownames of cells.

---

```
fluidigmSC_identifyExpOutliers
```

*An outlier detection function*

---

**Description**

An outlier detection function identifies cells with median expression below that of the bulk, reproduced from FluidigmSC package.

**Usage**

```
fluidigmSC_identifyExpOutliers(log2ex_data, expression_data_raw, threshold,
  step, fine_step, num_fine_test, pct_goodsample_threshold = 0.5,
  quantile_threshold = 0.95, low_quantile_threshold = 0.25,
  min_gene_number = 25, lod)
```

**Arguments**

log2ex_data	A data frame containing log <sub>2</sub> transformed expression values, with rownames of genes and colnames of cells.
expression_data_raw	A data frame containing raw expression values, with rownames of genes and colnames of cells.
threshold	A value in raw expression used as the starting threshold value.
step	An integer number indicating the increment of threshold value at each iteration.
fine_step	An integer number indicating the increment of threshold value at each iteration, at the refining stage.
num_fine_test	An integer number indicating the number of iteration of the refining stage.
pct_goodsample_threshold	A fraction value indicating the minimum percentage of samples on which the representative genes are detectable.
quantile_threshold	A probability of gene detection rate above which a sample is considered as good sample.

low_quantile_threshold	A probability of average gene expression value below which a sample is taken as an outlier.
min_gene_number	An integer indicating the minimum size of representative genes.
lod	A value of limit of detection in the unit of TPM/CPM/RPKM.

**Value**

A vector of character stating the IDs of outlier cells.

---

fluidigmSC\_isElementIgnoreCase  
*A gene finding function*

---

**Description**

A gene finding function looking for genes in the target set x from the source set y, reproduced from FluidigmSC package.

**Usage**

```
fluidigmSC_isElementIgnoreCase(x, y, ignore_case = TRUE)
```

**Arguments**

x	A vector of characters representing gene names (target genes).
y	A vector of characters representing gene names (source genes).
ignore_case	Boolean, if TRUE ignores letter case.

**Value**

A vector of characters representing gene names.

---

fluidigmSC\_readLinearExp

*An expression reading function*

---

### Description

An expression reading function which imports expression data from .txt file, and then computes log<sub>2</sub> transformed data, reproduced from FluidigmSC package.

### Usage

```
fluidigmSC_readLinearExp(exp_file = TRUE, lod = 1)
```

### Arguments

exp_file	Input file name in txt format, with rownames of cells and colnames of genes.
lod	A value of limit of detection in the unit of TPM/CPM/RPKM. It will be used as the starting value for outlier cell detection and the basis for removing scarce genes.

### Value

A list containing expression\_data\_raw(data frame), log<sub>2</sub>ex\_data(data frame), and log<sub>2</sub>ex\_avg\_data(data frame).

---

fluidigmSC\_removeGenesByLinearExpForAllType

*A gene trimming function*

---

### Description

A gene trimming function removes genes whose average expression value is below the log<sub>2</sub>(threshold), and also present in at least 10

### Usage

```
fluidigmSC_removeGenesByLinearExpForAllType(log2ex_data, log2ex_avg_data,
  threshold)
```

### Arguments

log2ex_data	A data frame containing log <sub>2</sub> transformed expression values, with rownames of genes and colnames of cells.
log2ex_avg_data	A data frame containing log <sub>2</sub> transformed average expression values for individual gene.
threshold	A limit of detection in the unit of TPM/CPM/RPKM.

**Value**

A vector of character containing gene names of those passed the filtering.

---

fluidigmSC\_removeGenesByLinearExpForAllType\_log2  
*A gene trimming function*

---

**Description**

A gene trimming function removes genes whose average expression value is below the  $\log_2(\text{threshold})$ ; reproduced from FluidigmSC package.

**Usage**

```
fluidigmSC_removeGenesByLinearExpForAllType_log2(log2ex_data, threshold)
```

**Arguments**

log2ex_data	A data frame containing $\log_2$ transformed expression values, with rownames of genes and colnames of cells.
threshold	A limit of detection in the unit of TPM/CPM/RPMK.

**Value**

A vector of character containing gene names of those passed the filtering.

---

monocle\_wrapper      *A wrapper function for Monocle sorting method*

---

**Description**

A wrapper function for Monocle sorting method

**Usage**

```
monocle_wrapper(log2_exp, expression_data_raw, lod = 1)
```

**Arguments**

log2_exp	An $\log_2$ transformed expression matrix containing n-rows of cells and m-cols of genes.
expression_data_raw	A data frame containing raw expression values, with rownames of cells and colnames of genes.
lod	A value of limit of detection in the unit of TPM/CPM/RPKM.

**Value**

A data frame containing single column of ordered sample IDs.

**Examples**

```
set.seed(15)
da <- iris[sample(150, 150, replace = FALSE), ]
rownames(da) <- paste0('spl_', seq(1, nrow(da)))
d <- da[, 1:4]
dl <- da[, 5, drop=FALSE]
#res <- monocle_wrapper(log2_exp = d, expression_data_raw = d)
#dl <- dl[match(res, rownames(dl)), ]
#annot <- data.frame(id = seq(1, length(res)), label=dl, stringsAsFactors = FALSE)
#ggplot(annot, aes(x=id, y=id, colour = label)) + geom_point() + theme_bw()
```

---

neighborhood\_sorting *A sorting function using the Neighborhood algorithm*

---

**Description**

A sorting function using the Neighborhood algorithm

**Usage**

```
neighborhood_sorting(d, weights_mat = NULL, max_iter = 100)
```

**Arguments**

d	A matrix containing n-by-n cell distance.
weights_mat	A weight matrix of size n-by-n.
max_iter	An integer number indicating the maximum number of iteration if sorting does not converge.

**Value**

A list containing ordering(a vector of re-ordered sequence) and cost(a numeric value).

---

neighborhood\_sortingcost

*A cost computation function for Neighborhood algorithm*

---

### Description

A cost computation function for Neighborhood algorithm

### Usage

```
neighborhood_sortingcost(expr = NULL, sigma_width = 1,  
  method = c("Euclidean", "Correlation", "eJaccard", "none"))
```

### Arguments

expr	An expression matrix containing n-rows of cells and m-cols of genes.
sigma_width	An integer number determining the degree of spread of the gaussian distribution which is used for computing weight matrix for Neighborhood sorting method.
method	A character string indicating the distance function.

### Value

A numeric value of sorting cost.

### Examples

```
set.seed(15)  
da <- iris[sample(150, 150, replace = FALSE), ]  
d <- da[,1:4]  
randomOrdering_cost <- neighborhood_sortingcost(d, method= 'Euclidean')  
randomOrdering_cost  
  
da <- iris  
d <- da[,1:4]  
properOrdering_cost <- neighborhood_sortingcost(d, method= 'Euclidean')  
properOrdering_cost
```

---

neighborhood\_sorting\_wrapper

*A wrapper function for Neighborhood sorting.*

---

### Description

A wrapper function for Neighborhood sorting as proposed in [Tsafirir et al. 2005].

**Usage**

```
neighborhood_sorting_wrapper(expr, sigma_width = 1, no_randomization = 10)
```

**Arguments**

`expr` An expression matrix containing n-rows of cells and m-cols of genes.

`sigma_width` An integer number determining the degree of spread of the gaussian distribution which is used for computing weight matrix for Neighborhood sorting method.

`no_randomization` An integer number indicating the number of repeated sorting, each of which uses a randomly selected initial cell ordering.

**Value**

A list containing `permutated.expr`(data frame) and `best.cost`(a numeric value).

---

`pca_gene_selection`      *Gene selection using PCA technique*

---

**Description**

Gene selection using PCA technique

**Usage**

```
pca_gene_selection(data)
```

**Arguments**

`data` A matrix of data.frame with row.name of cells, and col.name of genes

**Value**

a vector of the names of selected genes.

**Examples**

```
dir <- system.file('extdata', package='uSORT')
file <- list.files(dir, pattern='.txt$', full=TRUE)
exprs <- uSORT_preProcess(exprs_file = file)
exp_trimmed <- t(exprs$exprs_log_trimmed)
PCA_selected_genes <- pca_gene_selection(exp_trimmed)
```



---

Rwanderlust

*R implementation of wanderlust*

---

## Description

R implementation of wanderlust

## Usage

```
Rwanderlust(data, s, l = 15, k = 15, num_graphs = 1,  
             num_waypoints = 250, waypoints_seed = 123, flock_waypoints = 2,  
             metric = "euclidean", voting_scheme = "exponential",  
             band_sample = FALSE, partial_order = NULL, verbose = TRUE)
```

## Arguments

data	Input data matrix.
s	Starting point ID.
l	l nearest neighbours.
k	k nearest neighbours, $k < l$ .
num_graphs	Number of repeated graphs.
num_waypoints	Number of waypoints to guide the trajectory detection.
waypoints_seed	The seed for reproducing the results.
flock_waypoints	The number of times for flocking the waypoints, default is 2.
metric	Distance calculation metric for nearest neighbour detection.
voting_scheme	The scheme of voting.
band_sample	Boolean, if band the sample
partial_order	default NULL
verbose	Boolean, if print the details

## Value

a list containing Trajectory, Order, Waypoints

## Author(s)

Hao Chen

**Examples**

```

set.seed(15)
shuffled_iris <- iris[sample(150, 150, replace = FALSE), ]
data <- shuffled_iris[,1:4]
data_label <- shuffled_iris[,5]
wishbone <- Rwanderlust(data = data, num_waypoints = 100, waypoints_seed = 2)
pd1 <- data.frame(id = wishbone$Trajectory, label=data_label, stringsAsFactors = FALSE)
pd2 <- data.frame(id = seq_along(row.names(data)), label=data_label, stringsAsFactors = FALSE)
#ggplot(pd1, aes(x=id, y=id, colour = label)) + geom_point() + theme_bw()
#ggplot(pd2, aes(x=id, y=id, colour = label)) + geom_point() + theme_bw()

```

---

scattering\_quantification\_per\_gene

*An expression scattering measurement function*

---

**Description**

An expression scattering measurement function computes the level of scattering for individual genes along the cell ordering

**Usage**

```
scattering_quantification_per_gene(CDS = NULL)
```

**Arguments**

CDS                    a Monocle's CellDataSet object

**Value**

integer

**Author(s)**

MaiChan Lau

---

sorting\_method\_parameter\_GUI

*GUI for sorting method paramters*

---

**Description**

The parameters appeared on GUI are based on input method, this function is called by [uSORT\\_parameters\\_GUI](#). For internal use only.

**Usage**

```
sorting_method_parameter_GUI(method = c("autoSPIN", "sWanderlust", "monocle",
"Wanderlust", "SPIN", "none"))
```

**Arguments**

method            method name.

**Value**

a list of parameters.

**Author(s)**

Hao Chen

---

SPIN

*A wrapper function for SPIN sorting method*

---

**Description**

A wrapper function for SPIN method provides a R version of SPIN [Tsafrir et al. 2005].

**Usage**

```
SPIN(data, sorting_method = c("STS", "neighborhood"), sigma_width = 1)
```

**Arguments**

data            An log2 transformed expression matrix containing n-rows of cells and m-cols of genes.

sorting\_method   A character string indicating the choice of sorting method, i.e. 'STS' (Side-to-Side) or 'Neighborhood'.

sigma\_width     An integer number determining the degree of spread of the gaussian distribution which is used for computing weight matrix for Neighborhood sorting method.

**Value**

A data frame containing single column of ordered sample IDs.

**Examples**

```

set.seed(15)
da <- iris[sample(150, 150, replace = FALSE), ]
rownames(da) <- paste0('spl_', seq(1, nrow(da)))
d <- da[, 1:4]
d1 <- da[, 5, drop=FALSE]
res <- SPIN(data = d)
d1 <- d1[match(res$SampleID, rownames(d1)), ]
annot <- data.frame(id = seq(1, nrow(res)), label=d1, stringsAsFactors = FALSE)
#ggplot(annot, aes(x=id, y=id, colour = label)) + geom_point() + theme_bw()

```

---

STS\_sorting

*A sorting function using the Side-to-Side (STS) algorithm*


---

**Description**

A sorting function using the Side-to-Side (STS) algorithm

**Usage**

```
STS_sorting(d, max_iter = 10)
```

**Arguments**

**d** A matrix containing n-by-n cell distance.

**max\_iter** An integer number indicating the maximum number of iteration if sorting does not converge.

**Value**

A list containing ordering(a vector of re-ordered sequence) and cost(a numeric value).

---

STS\_sortingcost

*A cost computation function for Side-to-Side (STS) algorithm*


---

**Description**

A cost computation function for Side-to-Side (STS) algorithm

**Usage**

```

STS_sortingcost(expr = NULL, method = c("Euclidean", "Correlation",
    "eJaccard", "none"))

```

**Arguments**

`expr` An expression matrix containing n-rows of cells and m-cols of genes.  
`method` A character string indicating the distance function.

**Value**

A numeric value of sorting cost.

**Examples**

```
set.seed(15)
da <- iris[sample(150, 150, replace = FALSE), ]
d <- da[,1:4]
randomOrdering_cost <- STS_sortingcost(d, method= 'Euclidean')
randomOrdering_cost

da <- iris
d <- da[,1:4]
properOrdering_cost <- STS_sortingcost(d, method= 'Euclidean')
properOrdering_cost
```

---

STS\_sorting\_wrapper *A wrapper function for Side-to-Side (STS) sorting.*

---

**Description**

A wrapper function for Side-to-Side (STS) sorting as proposed in [Tsafrir et al. 2005].

**Usage**

```
STS_sorting_wrapper(expr, no_randomization = 10)
```

**Arguments**

`expr` An expression matrix containing n-rows of cells and m-cols of genes.  
`no_randomization` An integer number indicating the number of repeated sorting, each of which uses a randomly selected initial cell ordering.

**Value**

A list containing `permutated.expr`(data frame) and `best.cost`(a numeric value).

---

summed\_local\_variance *A summed local variance function*

---

### Description

A summed local variance function

### Usage

```
summed_local_variance(expr = NULL, alpha = NULL, data_type = "linear")
```

### Arguments

expr	An expression matrix containing n-rows of cells and m-cols of genes.
alpha	A fraction value indicating the size of window for local variance measurement.
data_type	A character string indicating the type of progression, i.e. 'linear' (strictly linear) or 'cyclical' (cyclically linear).

### Value

A numeric value of the summed local variance.

---

summed\_local\_variance\_cyclical  
*A summed local variance function for cyclical linear data type*

---

### Description

A summed local variance function for cyclical linear data type

### Usage

```
summed_local_variance_cyclical(d, alpha = 0.3)
```

### Arguments

d	A cell-to-cell distance matrix.
alpha	A fraction value indicating the size of window for local variance measurement.

### Value

A numeric value of the summed local variance.

---

summed\_local\_variance\_linear

*A summed local variance function for strictly linear data type*


---

**Description**

A summed local variance function for strictly linear data type

**Usage**

```
summed_local_variance_linear(d, alpha = 0.3)
```

**Arguments**

d	A cell-to-cell distance matrix.
alpha	A fraction value indicating the size of window for local variance measurement.

**Value**

A numeric value of the summed local variance.

---

sWanderlust

*sWanderlust*


---

**Description**

autoSPIN guided wanderlust. Specifically, we use autoSPIN to help find the starting point for wanderlust.

**Usage**

```
sWanderlust(data, data_type = c("linear", "cyclical"),
  SPIN_option = c("STS", "neighborhood"), alpha = 0.2, sigma_width = 1,
  diffusionmap_components = 4, l = 15, k = 15, num_waypoints = 150,
  flock_waypoints = 2, waypoints_seed = 2711)
```

**Arguments**

data	data Input data matrix.
data_type	The data type which guides the autoSPIN sorting, including linear, cyclical.
SPIN_option	SPIN contains two options including STS(default), neighborhood.
alpha	alpha parameter for autoSPIN, default is 0.2.
sigma_width	Sigma width parameter for SPIN, default is 1.

diffusionmap\_components      Number of components from diffusion map used for wanderlust analysis, default is 4.

l                                  Number of nearest neighbors, default is 15.

k                                  Number of nearest neighbors for repeating graphs, default is 15, should be less than or equal to l.

num\_waypoints      Number of waypoint used for wanderlust, default is 150.

flock\_waypoints                  The number of times for flocking the waypoints, default is 2.

waypoints\_seed      The seed for reproducing the results.

**Value**

a vector of the sorted order.

**Author(s)**

Hao Chen

**Examples**

```
set.seed(15)
shuffled_iris <- iris[sample(150, 150, replace = FALSE), ]
data <- shuffled_iris[,1:4]
data_label <- shuffled_iris[,5]
wishbone <- sWanderlust(data = data, num_waypoints = 100)
```

---

trajectory\_landmarks      *determining initial trajectory and landmarks*

---

**Description**

determining initial trajectory and landmarks

**Usage**

```
trajectory_landmarks(knn, data, s, partial_order = NULL, waypoints = 250,
  waypoints_seed = 123, metric = "euclidean", flock_waypoints = 2,
  band_sample = FALSE)
```

**Arguments**

knn                                  A sparse matrix of knn.

data                                  data.

s    The ID of starting point.

partial\_order      A vector of IDs specified as recommended waypoints, NULL to ignore.



waypoints	Either the number of waypoints, or specify the waypoint IDs.
waypoints_seed	Random sampling seed, for reproducible results.
metric	Distance calculation metric for nearest neighbour detection.
flock_waypoints	Iteration of using nearest points around waypoint to adjust its position.
band_sample	if give more chance to nearest neighbours of starting point in randomly waypoints selection.

**Value**

a list

---

uSORT	<i>uSORT: A self-refining ordering pipeline for gene selection</i>
-------	--

---

**Description**

This package is designed to uncover the intrinsic cell progression path from single-cell RNA-seq data.

The main function of uSORT-pacakge which provides a workflow of sorting scRNA-seq data.

**Usage**

```
uSORT(exprs_file, log_transform = TRUE, remove_outliers = TRUE,
      preliminary_sorting_method = c("autoSPIN", "sWanderlust", "monocle",
      "Wanderlust", "SPIN", "none"), refine_sorting_method = c("autoSPIN",
      "sWanderlust", "monocle", "Wanderlust", "SPIN", "none"),
      project_name = "uSORT", result_directory = getwd(), nCores = 1,
      save_results = TRUE, reproduce_seed = 1234,
      scattering_cutoff_prob = 0.75, driving_force_cutoff = NULL,
      qual_cutoff_featureSelection = 0.05, pre_data_type = c("linear",
      "cyclical"), pre_SPIN_option = c("STS", "neighborhood"),
      pre_SPIN_sigma_width = 1, pre_autoSPIN_alpha = 0.2,
      pre_autoSPIN_randomization = 20, pre_wanderlust_start_cell = NULL,
      pre_wanderlust_dfmap_components = 4, pre_wanderlust_l = 15,
      pre_wanderlust_num_waypoints = 150, pre_wanderlust_waypoints_seed = 2711,
      pre_wanderlust_flock_waypoints = 2, ref_data_type = c("linear",
      "cyclical"), ref_SPIN_option = c("STS", "neighborhood"),
      ref_SPIN_sigma_width = 1, ref_autoSPIN_alpha = 0.2,
      ref_autoSPIN_randomization = 20, ref_wanderlust_start_cell = NULL,
      ref_wanderlust_dfmap_components = 4, ref_wanderlust_l = 15,
      ref_wanderlust_num_waypoints = 150, ref_wanderlust_flock_waypoints = 2,
      ref_wanderlust_waypoints_seed = 2711)
```

**Arguments**

<code>exprs_file</code>	Input file name in txt format, with rownames of cells and colnames of genes.
<code>log_transform</code>	Boolean, if log transform the data.
<code>remove_outliers</code>	Boolean, if remove the outliers.
<code>preliminary_sorting_method</code>	Method name for preliminary sorting, including autoSPIN, sWanderlust, monocle, Wanderlust, SPIN, or none.
<code>refine_sorting_method</code>	Method name for refined sorting, including autoSPIN, sWanderlust, monocle, Wanderlust, SPIN, or none.
<code>project_name</code>	A character name as the prefix of the saved result file.
<code>result_directory</code>	The directory indicating where to save the results.
<code>nCores</code>	Number of cores that will be employed for drive gene selection (parallel computing), default is 1.
<code>save_results</code>	Boolean determining if save the results.
<code>reproduce_seed</code>	A seed used for reproducing the result.
<code>scattering_cutoff_prob</code>	Scattering cutoff value probability for gene selection, default 0.75.
<code>driving_force_cutoff</code>	Driving force cutoff value for gene selection, default NULL(automatically).
<code>qval_cutoff_featureSelection</code>	Q value cutoff for gene selection, default 0.05.
<code>pre_data_type</code>	The data type which guides the autoSPIN sorting, including linear, cyclical.
<code>pre_SPIN_option</code>	SPIN contains two options including STS(default), neighborhood.
<code>pre_SPIN_sigma_width</code>	Sigma width parameter for SPIN, default is 1.
<code>pre_autoSPIN_alpha</code>	alpha parameter for autoSPIN, default is 0.2.
<code>pre_autoSPIN_randomization</code>	Number of randomizations for autoSPIN, default is 20.
<code>pre_wanderlust_start_cell</code>	The name of starting cell for wanderlust, default is the first cell from the data.
<code>pre_wanderlust_dfmap_components</code>	Number of components from diffusion map used for wanderlust analysis, default is 4.
<code>pre_wanderlust_l</code>	Number of nearest neighbors used for wanderlust, default is 15.
<code>pre_wanderlust_num_waypoints</code>	Number of waypoint used for wanderlust, default is 150.
<code>pre_wanderlust_waypoints_seed</code>	The seed for reproducing the wanderlust results.

pre\_wanderlust\_flock\_waypoints  
The number of times for flocking the waypoints, default is 2.

ref\_data\_type The data type which guides the autoSPIN sorting, including linear, cyclical.

ref\_SPIN\_option  
SPIN contains two options including STS(default), neighborhood.

ref\_SPIN\_sigma\_width  
Sigma width parameter for SPIN, default is 1.

ref\_autoSPIN\_alpha  
alpha parameter for autoSPIN, default is 0.2.

ref\_autoSPIN\_randomization  
Number of randomizations for autoSPIN, default is 20.

ref\_wanderlust\_start\_cell  
The name of starting cell for wanderlust, default is the first cell from the data.

ref\_wanderlust\_dfmap\_components  
Number of components from diffusion map used for wanderlust analysis, default is 4.

ref\_wanderlust\_l  
Number of nearest neighbors used for wanderlust, default is 15.

ref\_wanderlust\_num\_waypoints  
Number of waypoint used for wanderlust, default is 150

ref\_wanderlust\_flock\_waypoints  
The number of times for flocking the waypoints, default is 2.

ref\_wanderlust\_waypoints\_seed  
The seed for reproducing the wanderlust results.

## Details

This package incorporates data pre-processing, preliminary PCA gene selection, preliminary cell ordering, feature selection, refined cell ordering, and post-analysis interpretation and visualization. The uSORT workflow can be implemented through calling the main function or the GUI. [uSORT](#).

## Value

results object (a list)

## See Also

[uSORT-package](#), [uSORT\\_GUI](#)

## Examples

```
dir <- system.file('extdata', package='uSORT')
file <- list.files(dir, pattern='.txt$', full=TRUE)
#remove the # symbol of the following codes to test
#uSORT_results <- uSORT(exprs_file = file, project_name = "test",
# preliminary_sorting_method = "autoSPIN",
# refine_sorting_method = "sWanderlust",
# save_results = FALSE)
```

uSORT\_GUI

*The user friendly GUI for uSORT-package*

---

**Description**

This GUI provides an easy way for applying the uSORT package.

**Usage**

```
uSORT_GUI()
```

**Value**

the GUI for uSORT-package

**Author(s)**

Hao Chen

**References**

<http://JinmiaoChenLab.github.io/uSORT/>

**See Also**

[uSORT-package](#), [uSORT](#)

**Examples**

```
interactive()  
#if(interactive()) uSORT_GUI() # remove the hash symbol to run
```

---

uSORT\_parameters\_GUI *The GUI for inputting paramters for uSORT*

---

**Description**

This is a function for generating the GUI for uSORT, it's called by [uSORT\\_GUI](#). For internal use only.

**Usage**

```
uSORT_parameters_GUI()
```

**Value**

a list of parameters.

**Author(s)**

Hao Chen

---

uSORT\_preProcess      *A data loading and pre-processing function*

---

**Description**

A data loading and pre-processing function which firstly identifies outlier cells and scarcely expressed genes.

**Usage**

```
uSORT_preProcess(exprs_file, log_transform = TRUE, remove_outliers = TRUE,  
                 lod = 1)
```

**Arguments**

`exprs_file`      Input file name in txt format, with rownames of cells and colnames of genes.

`log_transform`    Boolean, if TRUE log transform the data.

`remove_outliers`  
                    Boolean, if TRUE remove the outliers.

`lod`              A value of limit of detection in the unit of TPM/CPM/RPKM. It will be used as the starting value for outlier cell detection and the basis for removing scarce genes.

**Value**

A list containing `exprs_raw`(data frame) and `exprs_log_trimmed`(data.frame).

**Examples**

```
dir <- system.file('extdata', package='uSORT')  
file <- list.files(dir, pattern='.txt$', full=TRUE)  
exprs <- uSORT_preProcess(exprs_file = file)
```

---

uSORT\_sorting\_wrapper *wrapper of all available sorting methods in uSORT*

---

### Description

Sorting methods include autoSPIN, sWanderlust, monocle, Wanderlust, SPIN. Any of the sorting method can be called directly using this function.

### Usage

```
uSORT_sorting_wrapper(data, data_raw, method = c("autoSPIN", "sWanderlust",
  "monocle", "Wanderlust", "SPIN", "none"), data_type = c("linear",
  "cyclical"), SPIN_option = c("STS", "neighborhood"), SPIN_sigma_width = 1,
  autoSPIN_alpha = 0.2, autoSPIN_randomization = 20,
  wanderlust_start_cell = NULL, wanderlust_dfmap_components = 4,
  wanderlust_l = 15, wanderlust_num_waypoints = 150,
  wanderlust_waypoints_seed = 2711, wanderlust_flock_waypoints = 2)
```

### Arguments

data	Input preprocessed data matrix with row.name of cells and col.name of genes.
data_raw	Input raw data matrix with row.name of cells and col.name of genes, for monocle method.
method	The name of the sorting method to use, including autoSPIN, sWanderlust, monocle, Wanderlust, SPIN and none.
data_type	The type of the data, either linear or cyclical.
SPIN_option	The tuning option of SPIN, STS or neighborhood.
SPIN_sigma_width	Sigma width for SPIN.
autoSPIN_alpha	alpha for autoSPIN.
autoSPIN_randomization	Number of randomization for autoSPIN.
wanderlust_start_cell	The id of the starting cell for wanderlust.
wanderlust_dfmap_components	The number of components from diffusionmap for wanderlust.
wanderlust_l	The number of nearest neighbors used for wanderlust.
wanderlust_num_waypoints	The number of waypoints for wanderlust.
wanderlust_waypoints_seed	The seed for reproducible analysis.
wanderlust_flock_waypoints	The number of flock times for wanderlust.

**Value**

return the order of sorting results.

**Examples**

```
dir <- system.file('extdata', package='uSORT')
file <- list.files(dir, pattern='.txt$', full=TRUE)
exprs <- uSORT_preProcess(exprs_file = file)
exp_trimmed <- t(exprs$exprs_log_trimmed)
PCA_selected_genes <- pca_gene_selection(exp_trimmed)
exp_PCA_genes <- exp_trimmed[, PCA_selected_genes]
#order <- uSORT_sorting_wrapper(data = exp_PCA_genes, method = 'autoSPIN')
```

---

uSORT\_write\_results     *Results parsing for uSORT*

---

**Description**

Save result object into a RData file. Save cell to cell distance heatmap for both preliminary and refined results. Create plot of driver gene profiles on final ordering using heatmap.

**Usage**

```
uSORT_write_results(uSORT_results, project_name, result_directory)
```

**Arguments**

uSORT\_results     Result object from uSort function, a list.  
project\_name     A prefix for the saving files.  
result\_directory     The path where to save the results.

**Value**

save the results.

**Examples**

```
dir <- system.file('extdata', package='uSORT')
file <- list.files(dir, pattern='.txt$', full=TRUE)
#remove the # symbol of the following codes to test
#uSORT_results <- uSORT(exprs_file = file,
# project_name = 'test',
# preliminary_sorting_method = 'autoSPIN',
# refine_sorting_method = 'sWanderlust',
# save_results = FALSE)
#uSORT_write_results(uSORT_results,
# project_name = 'test',
# result_directory = getwd())
```

---

variability\_per\_gene *A utility function for scattering\_quantification\_per\_gene*

---

### Description

A utility function for scattering\_quantification\_per\_gene which computes the degree of scattering for single gene, whereby the value is computed by summing over the local values of smaller local windows

### Usage

```
variability_per_gene(logExp = NULL, min_expr = 0.1,
  window_size_perct = 0.1, nonZeroExpr_perct = 0.1)
```

### Arguments

logExp	a log-scale expression vector of a gene
min_expr	a minimum expression value
window_size_perct	a window size (in dispersion level)
nonZeroExpr_perct	a minimum amount of cells (in expression, otherwise the associated window will be assigned to 0 dispersion value)

### Value

integer

### Author(s)

MaiChan Lau

---

wanderlust\_wrapper *a wrapper of wanderlust for sWanderlust*

---

### Description

a wrapper of wanderlust for sWanderlust

### Usage

```
wanderlust_wrapper(data, s, diffusionmap_components = 4, l = 15, k = 15,
  num_graphs = 1, num_waypoints = 150, waypoints_seed = 123,
  flock_waypoints = 2)
```



**Arguments**

<code>data</code>	Input data matrix.
<code>s</code>	The ID of starting point.
<code>diffusionmap_components</code>	Number of components from diffusion map used for wanderlust analysis, default is 4.
<code>l</code>	Number of nearest neighbors, default is 15.
<code>k</code>	Number of nearest neighbors for repeating graphs, default is 15, should be less than or equal to <code>l</code> .
<code>num_graphs</code>	Number of repeated graphs.
<code>num_waypoints</code>	Number of waypoint used for wanderlust, default is 150.
<code>waypoints_seed</code>	The seed for reproducing the results.
<code>flock_waypoints</code>	The number of times for flocking the waypoints, default is 2.

**Value**

sorted order.

**Author(s)**

Hao Chen

# Index

autoSPIN, [3](#)

clusterGenes1, [4](#)  
compareModels1, [5](#)

diff\_test\_helper1, [6](#)  
differentialGeneTest1, [5](#)  
distance.function, [7](#)  
driving\_force\_gene\_selection, [7](#)

elbow\_detection, [8](#)  
EXP\_to\_CellDataSet, [9](#)

fluidigmSC\_analyzeGeneDetection, [9](#)  
fluidigmSC\_identifyExpOutliers, [10](#)  
fluidigmSC\_isElementIgnoreCase, [11](#)  
fluidigmSC\_readLinearExp, [12](#)  
fluidigmSC\_removeGenesByLinearExpForAllType,  
[12](#)  
fluidigmSC\_removeGenesByLinearExpForAllType\_log2,  
[13](#)

monocle\_wrapper, [13](#)

neighborhood\_sorting, [14](#)  
neighborhood\_sorting\_wrapper, [15](#)  
neighborhood\_sortingcost, [15](#)

pca\_gene\_selection, [16](#)

Rwanderlust, [17](#)

scattering\_quantification\_per\_gene, [18](#)  
sorting\_method\_parameter\_GUI, [18](#)  
SPIN, [19](#)  
STS\_sorting, [20](#)  
STS\_sorting\_wrapper, [21](#)  
STS\_sortingcost, [20](#)  
summed\_local\_variance, [22](#)  
summed\_local\_variance\_cyclical, [22](#)  
summed\_local\_variance\_linear, [23](#)

sWanderlust, [23](#)

trajectory\_landmarks, [24](#)

uSORT, [25](#), [27](#), [28](#)  
uSORT-package (uSORT), [25](#)  
uSORT\_GUI, [27](#), [28](#), [28](#)  
uSORT\_parameters\_GUI, [18](#), [28](#)  
uSORT\_preProcess, [29](#)  
uSORT\_sorting\_wrapper, [30](#)  
uSORT\_write\_results, [31](#)

variability\_per\_gene, [32](#)

wanderlust\_wrapper, [32](#)