

Package ‘harmony’

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Title Fast, Sensitive, and Accurate Integration of Single Cell Data

Version 1.2.4

Description Implementation of the Harmony algorithm for single cell integration, described in Korsunsky et al <[doi:10.1038/s41592-019-0619-0](https://doi.org/10.1038/s41592-019-0619-0)>. Package includes a standalone Harmony function and interfaces to external frameworks.

URL <https://github.com/immunogenomics/harmony>

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Encoding UTF-8

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cell_lines	<i>List of metadata table and scaled PCs matrix</i>
------------	---

Description

List of metadata table and scaled PCs matrix

Usage

cell_lines

Format

: meta_data: data.table of 9478 rows with defining dataset and cell_type scaled_pcs: data.table of 9478 rows (cells) and 20 columns (PCs)

Source

<https://www.10xgenomics.com>

cell_lines_small	<i>Same as cell_lines but smaller (300 cells).</i>
------------------	--

Description

Same as cell_lines but smaller (300 cells).

Usage

```
cell_lines_small
```

Format

An object of class list of length 2.

Source

<https://www.10xgenomics.com>

harmony	<i>Harmony: fast, accurate, and robust single cell integration.</i>
---------	---

Description

Algorithm for single cell integration.

Usage

?RunHarmony to run Harmony on cell embeddings matrix, Seurat or SingleCellExperiment objects.

Useful links

1. Report bugs at <https://github.com/immunogenomics/harmony/issues>
2. Read the manuscript [doi:10.1038/s4159201906190](https://doi.org/10.1038/s4159201906190)

HarmonyMatrix

*A proxy call to [RunHarmony\(\)](#). *Deprecated.**

Description

Maintain name backwards compatibility with version 0 of harmony. However, API is not backwards compatible with version 0. This function will be deprecated in later versions of Harmony.

Usage

```
HarmonyMatrix(...)
```

Arguments

... Arguments passed on to [RunHarmony.default](#)

`data_mat` Matrix of cell embeddings. Cells can be rows or columns and will be inferred by the rows of `meta_data`.

`meta_data` Either (1) Dataframe with variables to integrate or (2) vector with labels.

`vars_use` If `meta_data` is dataframe, this defined which variable(s) to remove (character vector).

`theta` Diversity clustering penalty parameter. Specify for each variable in `vars_use`. Default `theta=2`. `theta=0` does not encourage any diversity. Larger values of `theta` result in more diverse clusters.

`sigma` Width of soft kmeans clusters. Default `sigma=0.1`. `Sigma` scales the distance from a cell to cluster centroids. Larger values of `sigma` result in cells assigned to more clusters. Smaller values of `sigma` make soft kmeans cluster approach hard clustering.

`lambda` Ridge regression penalty. Default `lambda=1`. Bigger values protect against over correction. If several covariates are specified, then `lambda` can also be a vector which needs to be equal length with the number of variables to be corrected. In this scenario, each covariate level group will be assigned the scalars specified by the user. If set to `NULL`, harmony will start `lambda` estimation mode to determine `lambdas` automatically and try to minimize overcorrection (Use with caution still in beta testing).

`nclust` Number of clusters in model. `nclust=1` equivalent to simple linear regression.

`max_iter` Maximum number of rounds to run Harmony. One round of Harmony involves one clustering and one correction step.

`early_stop` Enable early stopping for harmony. The harmonization process will stop when the change of objective function between corrections drops below `1e-4`

`ncores` Number of processors to be used for math operations when optimized BLAS is available. If BLAS is not supporting multithreaded then this option has no effect. By default, `ncore=1` which runs as a single-threaded

process. Although Harmony supports multiple cores, it is not optimized for multithreading. Increase this number for large datasets iff single-core performance is not adequate.

`plot_convergence` Whether to print the convergence plot of the clustering objective function. TRUE to plot, FALSE to suppress. This can be useful for debugging.

`return_object` (Advanced Usage) Whether to return the Harmony object or only the corrected PCA embeddings.

`verbose` Whether to print progress messages. TRUE to print, FALSE to suppress.

`.options` Setting advanced parameters of RunHarmony. This must be the result from a call to 'harmony_options'. See `harmony_options` for parameters not listed above and more details.

harmony_options

Set advanced parameters for RunHarmony

Description

Set advanced parameters for RunHarmony

Usage

```
harmony_options(
  alpha = 0.2,
  tau = 0,
  block.size = 0.05,
  max.iter.cluster = 20,
  epsilon.cluster = 0.001,
  epsilon.harmony = 0.01
)
```

Arguments

<code>alpha</code>	When setting <code>lambda = NULL</code> and use lambda estimation mode, lambda would be determined by the expected number of cells assuming independence between batches and clusters. i.e., $\lambda = \alpha * \text{expected number of cells}$, default 0.2 and alpha should be $0 < \alpha < 1$
<code>tau</code>	Protection against overclustering small datasets with large ones. 'tau' is the expected number of cells per cluster.
<code>block.size</code>	What proportion of cells to update during clustering. Between 0 to 1, default 0.05. Larger values may be faster but less accurate.
<code>max.iter.cluster</code>	Maximum number of rounds to run clustering at each round of Harmony.

epsilon.cluster
Convergence tolerance for clustering round of Harmony. Set to -Inf to never stop early.

epsilon.harmony
Convergence tolerance for Harmony. Set to -Inf to never stop early. When 'epsilon.harmony' is set to not NULL, then user-supplied values of 'early_stop' is ignored.

Value

Return a list for '.options' argument of 'RunHarmony'

Examples

```
## If want to set max.iter.cluster to be 100, do
## Not run:
RunHarmony(data_meta, meta_data, vars_use,
            .options = harmony_options(max.iter.cluster = 100))

## End(Not run)
```

moe_ridge_get_betas *Get beta Utility*

Description

Utility function to get ridge regression coefficients from trained Harmony object

Usage

```
moe_ridge_get_betas(harmonyObj)
```

Arguments

harmonyObj Trained harmony object. Get this by running RunHarmony function with return_object=TRUE.

Value

Returns nothing, modifies object in place.

pbmc.ctrl	<i>Gene expression data of control PBMC from Kang et al. 2017. This contains a sample of 1000 cells from that condition and is used for the Seurat Vignette.</i>
-----------	--

Description

Gene expression data of control PBMC from Kang et al. 2017. This contains a sample of 1000 cells from that condition and is used for the Seurat Vignette.

Usage

```
pbmc.ctrl
```

Format

An object of class `dgMatrix` with 9015 rows and 1000 columns.

Source

[doi:10.1038/nbt.4042](https://doi.org/10.1038/nbt.4042)

pbmc.stim	<i>Gene expression data of stimulated PBMC from Kang et al. 2017. This contains a sample of 1000 cells from that condition and is used for the Seurat Vignette.</i>
-----------	---

Description

Gene expression data of stimulated PBMC from Kang et al. 2017. This contains a sample of 1000 cells from that condition and is used for the Seurat Vignette.

Usage

```
pbmc.stim
```

Format

An object of class `dgMatrix` with 9015 rows and 1000 columns.

Source

[doi:10.1038/nbt.4042](https://doi.org/10.1038/nbt.4042)

RunHarmony	<i>Generic function that runs the harmony algorithm on single-cell genomics cell embeddings.</i>
------------	--

Description

RunHarmony is generic function that runs the main Harmony algorithm. If working with single cell R objects, please refer to the documentation of the appropriate generic API: ([RunHarmony.Seurat\(\)](#) or [RunHarmony.SingleCellExperiment\(\)](#)). If users work with other forms of cell embeddings, they can pass them directly to harmony using [RunHarmony.default\(\)](#) API. All the function arguments listed here are common in all RunHarmony interfaces.

Usage

```
RunHarmony(...)
```

Arguments

... Arguments passed on to [RunHarmony.default](#)

theta Diversity clustering penalty parameter. Specify for each variable in `vars_use`. Default `theta=2`. `theta=0` does not encourage any diversity. Larger values of `theta` result in more diverse clusters.

sigma Width of soft kmeans clusters. Default `sigma=0.1`. Sigma scales the distance from a cell to cluster centroids. Larger values of `sigma` result in cells assigned to more clusters. Smaller values of `sigma` make soft kmeans cluster approach hard clustering.

lambda Ridge regression penalty. Default `lambda=1`. Bigger values protect against over correction. If several covariates are specified, then `lambda` can also be a vector which needs to be equal length with the number of variables to be corrected. In this scenario, each covariate level group will be assigned the scalars specified by the user. If set to `NULL`, harmony will start lambda estimation mode to determine lambdas automatically and try to minimize overcorrection (Use with caution still in beta testing).

nclust Number of clusters in model. `nclust=1` equivalent to simple linear regression.

max_iter Maximum number of rounds to run Harmony. One round of Harmony involves one clustering and one correction step.

early_stop Enable early stopping for harmony. The harmonization process will stop when the change of objective function between corrections drops below $1e-4$

ncores Number of processors to be used for math operations when optimized BLAS is available. If BLAS is not supporting multithreaded then this option has no effect. By default, `ncore=1` which runs as a single-threaded process. Although Harmony supports multiple cores, it is not optimized for multithreading. Increase this number for large datasets iff single-core performance is not adequate.

- plot_convergence Whether to print the convergence plot of the clustering objective function. TRUE to plot, FALSE to suppress. This can be useful for debugging.
- verbose Whether to print progress messages. TRUE to print, FALSE to suppress.
- .options Setting advanced parameters of RunHarmony. This must be the result from a call to 'harmony_options'. See `harmony_options` for parameters not listed above and more details.

Value

If used with single-cell objects, it will return the updated single-cell object. For standalone operation, it returns the corrected cell embeddings or the R6 harmony object (see `RunHarmony.default()`).

See Also

Other RunHarmony: `RunHarmony.Seurat()`, `RunHarmony.SingleCellExperiment()`, `RunHarmony.default()`

`RunHarmony.default` *This is the primary harmony interface.*

Description

Use this generic with a cell embeddings matrix, a metadata table and a categorical covariate to run the Harmony algorithm directly on cell embedding matrix.

Usage

```
## Default S3 method:
RunHarmony(
  data_mat,
  meta_data,
  vars_use,
  theta = NULL,
  sigma = 0.1,
  lambda = 1,
  nclust = NULL,
  max_iter = 10,
  early_stop = TRUE,
  ncores = 1,
  plot_convergence = FALSE,
  return_object = FALSE,
  verbose = TRUE,
  .options = harmony_options(),
  ...
)
```

Arguments

data_mat	Matrix of cell embeddings. Cells can be rows or columns and will be inferred by the rows of meta_data.
meta_data	Either (1) Dataframe with variables to integrate or (2) vector with labels.
vars_use	If meta_data is dataframe, this defined which variable(s) to remove (character vector).
theta	Diversity clustering penalty parameter. Specify for each variable in vars_use Default theta=2. theta=0 does not encourage any diversity. Larger values of theta result in more diverse clusters.
sigma	Width of soft kmeans clusters. Default sigma=0.1. Sigma scales the distance from a cell to cluster centroids. Larger values of sigma result in cells assigned to more clusters. Smaller values of sigma make soft kmeans cluster approach hard clustering.
lambda	Ridge regression penalty. Default lambda=1. Bigger values protect against over correction. If several covariates are specified, then lambda can also be a vector which needs to be equal length with the number of variables to be corrected. In this scenario, each covariate level group will be assigned the scalars specified by the user. If set to NULL, harmony will start lambda estimation mode to determine lambdas automatically and try to minimize overcorrection (Use with caution still in beta testing).
nclust	Number of clusters in model. nclust=1 equivalent to simple linear regression.
max_iter	Maximum number of rounds to run Harmony. One round of Harmony involves one clustering and one correction step.
early_stop	Enable early stopping for harmony. The harmonization process will stop when the change of objective function between corrections drops below 1e-4
ncores	Number of processors to be used for math operations when optimized BLAS is available. If BLAS is not supporting multithreaded then this option has no effect. By default, ncore=1 which runs as a single-threaded process. Although Harmony supports multiple cores, it is not optimized for multithreading. Increase this number for large datasets iff single-core performance is not adequate.
plot_convergence	Whether to print the convergence plot of the clustering objective function. TRUE to plot, FALSE to suppress. This can be useful for debugging.
return_object	(Advanced Usage) Whether to return the Harmony object or only the corrected PCA embeddings.
verbose	Whether to print progress messages. TRUE to print, FALSE to suppress.
.options	Setting advanced parameters of RunHarmony. This must be the result from a call to 'harmony_options'. See <code>harmony_options</code> for parameters not listed above and more details.
...	other parameters that are not part of the API

Value

By default, matrix with corrected PCA embeddings. If return_object is TRUE, returns the full Harmony object (R6 reference class type).

See Also

Other RunHarmony: [RunHarmony.Seurat\(\)](#), [RunHarmony.SingleCellExperiment\(\)](#), [RunHarmony\(\)](#)

Examples

```
## By default, Harmony inputs a cell embedding matrix
## Not run:
harmony_embeddings <- RunHarmony(cell_embeddings, meta_data, 'dataset')

## End(Not run)

## If PCA is the input, the PCs need to be scaled
data(cell_lines_small)
pca_matrix <- cell_lines_small$scaled_pcs
meta_data <- cell_lines_small$meta_data
harmony_embeddings <- RunHarmony(pca_matrix, meta_data, 'dataset')

## Output is a matrix of corrected PC embeddings
dim(harmony_embeddings)
harmony_embeddings[seq_len(5), seq_len(5)]

## Finally, we can return an object with all the underlying data structures
harmony_object <- RunHarmony(pca_matrix, meta_data, 'dataset', return_object=TRUE)
dim(harmony_object$Y) ## cluster centroids
dim(harmony_object$R) ## soft cluster assignment
dim(harmony_object$Z_corr) ## corrected PCA embeddings
head(harmony_object$0) ## batch by cluster co-occurrence matrix
```

RunHarmony.Seurat *Applies harmony on a Seurat object cell embedding.*

Description

Applies harmony on a Seurat object cell embedding.

Usage

```
## S3 method for class 'Seurat'
RunHarmony(
  object,
  group.by.vars,
  reduction.use = "pca",
  dims.use = NULL,
  reduction.save = "harmony",
  project.dim = TRUE,
  ...
)
```

Arguments

<code>object</code>	the Seurat object. It needs to have the appropriate slot of cell embeddings pre-computed.
<code>group.by.vars</code>	the name(s) of covariates that harmony will remove its effect on the data.
<code>reduction.use</code>	Name of dimension reduction to use. Default is <code>pca</code> .
<code>dims.use</code>	indices of the cell embedding features to be used
<code>reduction.save</code>	the name of the new slot that is going to be created by harmony. By default, <code>harmony</code> .
<code>project.dim</code>	Project dimension reduction loadings. Default <code>TRUE</code> .
<code>...</code>	Arguments passed on to <code>RunHarmony.default</code>
<code>theta</code>	Diversity clustering penalty parameter. Specify for each variable in <code>vars_use</code> . Default <code>theta=2</code> . <code>theta=0</code> does not encourage any diversity. Larger values of <code>theta</code> result in more diverse clusters.
<code>sigma</code>	Width of soft kmeans clusters. Default <code>sigma=0.1</code> . <code>Sigma</code> scales the distance from a cell to cluster centroids. Larger values of <code>sigma</code> result in cells assigned to more clusters. Smaller values of <code>sigma</code> make soft kmeans cluster approach hard clustering.
<code>lambda</code>	Ridge regression penalty. Default <code>lambda=1</code> . Bigger values protect against over correction. If several covariates are specified, then <code>lambda</code> can also be a vector which needs to be equal length with the number of variables to be corrected. In this scenario, each covariate level group will be assigned the scalars specified by the user. If set to <code>NULL</code> , harmony will start <code>lambda</code> estimation mode to determine <code>lambdas</code> automatically and try to minimize overcorrection (Use with caution still in beta testing).
<code>nclust</code>	Number of clusters in model. <code>nclust=1</code> equivalent to simple linear regression.
<code>max_iter</code>	Maximum number of rounds to run Harmony. One round of Harmony involves one clustering and one correction step.
<code>early_stop</code>	Enable early stopping for harmony. The harmonization process will stop when the change of objective function between corrections drops below $1e-4$
<code>ncores</code>	Number of processors to be used for math operations when optimized BLAS is available. If BLAS is not supporting multithreaded then this option has no effect. By default, <code>ncore=1</code> which runs as a single-threaded process. Although Harmony supports multiple cores, it is not optimized for multithreading. Increase this number for large datasets iff single-core performance is not adequate.
<code>plot_convergence</code>	Whether to print the convergence plot of the clustering objective function. <code>TRUE</code> to plot, <code>FALSE</code> to suppress. This can be useful for debugging.
<code>verbose</code>	Whether to print progress messages. <code>TRUE</code> to print, <code>FALSE</code> to suppress.
<code>.options</code>	Setting advanced parameters of RunHarmony. This must be the result from a call to <code>'harmony_options'</code> . See <code>¿harmony_options'</code> for parameters not listed above and more details.

Value

Seurat object. Harmony dimensions placed into a new slot in the Seurat object according to the reduction.save. For downstream Seurat analyses, use reduction='harmony'.

See Also

Other RunHarmony: [RunHarmony.SingleCellExperiment\(\)](#), [RunHarmony.default\(\)](#), [RunHarmony\(\)](#)

Examples

```
## Not run:
## seu is a Seurat single-Cell R object
seu <- RunHarmony(seu, "donor_id")

## End(Not run)
```

RunHarmony.SingleCellExperiment

Applies harmony on PCA cell embeddings of a SingleCellExperiment.

Description

Applies harmony on PCA cell embeddings of a SingleCellExperiment.

Usage

```
## S3 method for class 'SingleCellExperiment'
RunHarmony(
  object,
  group.by.vars,
  dims.use = NULL,
  verbose = TRUE,
  reduction.save = "HARMONY",
  ...
)
```

Arguments

object	SingleCellExperiment with the PCA reducedDim cell embeddings populated
group.by.vars	the name(s) of covariates that harmony will remove its effect on the data.
dims.use	a vector of indices that allows only selected cell embeddings features to be used.
verbose	enable verbosity
reduction.save	the name of the new slot that is going to be created by harmony. By default, HARMONY.
...	Arguments passed on to RunHarmony.default

- theta** Diversity clustering penalty parameter. Specify for each variable in `vars_use`. Default `theta=2`. `theta=0` does not encourage any diversity. Larger values of `theta` result in more diverse clusters.
- sigma** Width of soft kmeans clusters. Default `sigma=0.1`. Sigma scales the distance from a cell to cluster centroids. Larger values of `sigma` result in cells assigned to more clusters. Smaller values of `sigma` make soft kmeans cluster approach hard clustering.
- lambda** Ridge regression penalty. Default `lambda=1`. Bigger values protect against over correction. If several covariates are specified, then `lambda` can also be a vector which needs to be equal length with the number of variables to be corrected. In this scenario, each covariate level group will be assigned the scalars specified by the user. If set to `NULL`, harmony will start `lambda` estimation mode to determine `lambdas` automatically and try to minimize overcorrection (Use with caution still in beta testing).
- nclust** Number of clusters in model. `nclust=1` equivalent to simple linear regression.
- max_iter** Maximum number of rounds to run Harmony. One round of Harmony involves one clustering and one correction step.
- early_stop** Enable early stopping for harmony. The harmonization process will stop when the change of objective function between corrections drops below `1e-4`.
- ncores** Number of processors to be used for math operations when optimized BLAS is available. If BLAS is not supporting multithreaded then this option has no effect. By default, `ncore=1` which runs as a single-threaded process. Although Harmony supports multiple cores, it is not optimized for multithreading. Increase this number for large datasets iff single-core performance is not adequate.
- plot_convergence** Whether to print the convergence plot of the clustering objective function. `TRUE` to plot, `FALSE` to suppress. This can be useful for debugging.
- .options** Setting advanced parameters of RunHarmony. This must be the result from a call to `'harmony_options'`. See `¿harmony_options'` for parameters not listed above and more details.

Value

SingleCellExperiment object. After running RunHarmony, the corrected cell embeddings can be accessed with `reducedDim(object, "Harmony")`.

See Also

Other RunHarmony: [RunHarmony.Seurat\(\)](#), [RunHarmony.default\(\)](#), [RunHarmony\(\)](#)

Examples

```
## Not run:
## sce is a SingleCellExperiment R object
sce <- RunHarmony(sce, "donor_id")
```

```
## End(Not run)
```

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