

# Package ‘attract’

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**Type** Package

**Title** Methods to Find the Gene Expression Modules that Represent the Drivers of Kauffman's Attractor Landscape

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**Description** This package contains the functions to find the gene expression modules that represent the drivers of Kauffman's attractor landscape. The modules are the core attractor pathways that discriminate between different cell types of groups of interest. Each pathway has a set of synexpression groups, which show transcriptionally-coordinated changes in gene expression.

**License** LGPL (>= 2.0)

**Collate** classdef.R findAttractorsStep.R removeFlatGenesStep.R findSynexprsStep.R findFuncEnrichAndCorr.R

**Depends** R (>= 2.10.0), AnnotationDbi, KEGG.db, limma, cluster,GOstats, graphics, methods, stats

**Imports** Biobase, AnnotationDbi, KEGG.db, limma, cluster, GOstats,graphics, methods, stats

**Suggests** illuminaHumanv1.db

**biocViews** Statistics, GeneExpression

## R topics documented:

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attract-package	<i>Methods to find the Gene Expression Modules that Represent the Drivers of Kauffman's Attractor Landscape</i>
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### Description

This package contains functions used to determine the gene expression modules that represent the drivers of Kauffman's attractor landscape.

### Details

Package:	attract
Type:	Package
Version:	1.0
Date:	2010-01-21
License:	
LazyLoad:	yes

The method can be summarized in the following key steps: (1) Determine core KEGG pathways that discriminate the most strongly between celltypes or experimental groups of interest (see `findAttractors`). (2) Find the different synexpression groups that are present within a core attractor pathway (see `findSynexprs`). (3) Find sets of genes that show highly similar profiles to the synexpression groups within an attractor pathway module (see `findCorrPartners`). (4) Test for functional enrichment for each of the synexpression groups to detect any potentially shared biological themes (see `calcFuncSynexprs`).

### Author(s)

Jessica Mar <jess@jimmy.harvard.edu>

## References

Kauffman S. 2004. A proposal for using the ensemble approach to understand genetic regulatory networks. *J Theor Biol.* 230:581. Mar JC, Wells CA, Quackenbush J. 2010. Identifying Gene Expression Modules that Represent the Drivers of Kauffman's Attractor Landscape. To Appear. Müller F et al. 2008. Regulatory networks define phenotypic classes of human stem cell lines. *Nature.* 455(7211): 401. Mar JC, Wells CA, Quackenbush J. 2010. Defining an Informativeness Metric for Clustering Gene Expression Data. To Appear.

## Examples

```
## Not run:
data(subset.loring.eset)
attractor.states <- findAttractors(subset.loring.eset, "celltype", nperm=10, annotation="illuminaHumanv1.db")
remove.these.genes <- removeFlatGenes(subset.loring.eset, "celltype", contrasts=NULL, limma.cutoff=0.05)
mapk.syn <- findSynexprs("04010", attractor.states, remove.these.genes)
mapk.cor <- findCorrPartners(mapk.syn, subset.loring.eset, remove.these.genes)
mapk.func <- calcFuncSynexprs(mapk.syn, attractor.states, "CC", annotation="illuminaHumanv1.db")

## End(Not run)
```

---

AttractorModuleSet-class

*Class AttractorModuleSet*

---

## Description

This is a class representation for storing the output of the `findAttractors` function.

## Objects from the Class

Objects are output by the function `findAttractors`. Objects can also be created by using `new("AttractorModuleSet", ...)`.

## Slots

`eSet`: ExpressionSet which primarily stores the expression data and the phenotype/sample data sets.

`cellTypeTag`: character string of the tag which stores the group membership information for the samples. Must be a column name of the data frame `pData(eset)`.

`incidenceMatrix`: incidence matrix used as input to GSEAIm.

`rankedPathways`: Data frame of significantly enriched pathways, ranked first by significance and then by size.

## Methods

No methods have yet been defined with class "AttractorModuleSet" in the signature.

**Note**

This class is better describe in the vignette.

**Author(s)**

Jessica Mar <jess@jimmy.harvard.edu>

**Examples**

```
## Not run:
new.attractmodule <- new("AttractorModuleSet", eSet=new("ExpressionSet"), cellTypeTag=character(1), incidenceMat

## End(Not run)
```

---

calcFuncSynexprs      *Functional enrichmental analysis for a set of synexpression groups.*

---

**Description**

This function performs functional enrichment for a given set of synexpression groups.

**Usage**

```
calcFuncSynexprs(mySynExpressionSet, myAttractorModuleSet, ontology = "BP", min.pvalue = 0.05, min.pwa
```

**Arguments**

mySynExpressionSet	SynExpressionSet object.
myAttractorModuleSet	AttractorModuleSet object.
ontology	character string specifying which GO ontology to use, either "MF", "BP", or "CC"; defaults to "BP".
min.pvalue	numeric value specifying adjusted P-value cut-off to use, categories with P-values <= min.pvalue will be reported.
min.pwaysize	integer specifying minimum size of the pathway or category to consider for enrichment analysis.
annotation	character string specifying the annotation package that corresponds to the chip platform the data was generated from.
...	additional arguments.

**Details**

This function performs a functional enrichment analysis on each synexpression group using the hyperGTest from the G0stats package. P-values are adjusted using the Benjamini-Hochberg correction method. Results are returned only if they satisfy the minimum P-value level, as specified by the min.pvalue argument.

**Value**

A list object.

**Author(s)**

Jessica Mar

**References**

Falcon, S. and R. Gentleman, Using GOSTats to test gene lists for GO term association. *Bioinformatics*, 2007. 23(2): p. 257-8.

**Examples**

```
## Not run:
data(subset.loring.eset)
attractor.states <- findAttractors(subset.loring.eset, "celltype", nperm=10, annotation="illuminaHumanv1.db")
remove.these.genes <- removeFlatGenes(subset.loring.eset, "celltype", contrasts=NULL, limma.cutoff=0.05)
mapk.syn <- findSynexprs("04010", attractor.states, remove.these.genes)
mapk.func <- calcFuncSynexprs(mapk.syn, attractor.states, "CC", annotation="illuminaHumanv1.db")

## End(Not run)
```

---

calcInform

*Function calculates the informativeness metric (average MSS) for a set of cluster assignments.*

---

**Description**

Function calculates the informativeness metric (average MSS) for a set of cluster assignments.

**Usage**

```
calcInform(exprs.dat, cl, class.vector)
```

**Arguments**

exprs.dat      a matrix of gene expression values.  
cl              a vector of cluster assignments.  
class.vector   a vector specifying the group membership of the samples.

**Details**

This function is also called internally by findSynexprs.

**Value**

A numeric value representing the average MSS value (informativeness metric) for a set of cluster assignments. For an informative cluster, the RSS values should be very small relative to those produced by the informativeness metric (the MSS values).

**Author(s)**

Jessica Mar

**References**

Mar, J., C. Wells, and J. Quackenbush, Defining an Informativeness Metric for Clustering Gene Expression Data. to appear, 2010.

**Examples**

```
## Not run:
library(cluster)
data(subset.loring.eset)
clustObj <- agnes(as.dist(1-t(cor(exprs(subset.loring.eset)))))
cinform.vals <- NULL
for( i in 1:10 ){
  cinform.vals <- c(cinform.vals, calcInform(exprs(subset.loring.eset), cutree(clustObj,i), pData(subset.loring.eset)))
}
k <- (1:10)[cinform.vals==max(cinform.vals)] # gives the optimal number of clusters

## End(Not run)
```

---

calcModfstat

*Function calculates a modified F-statistic for a set of cluster assignments.*

---

**Description**

Function calculates a modified F-statistic for a set of cluster assignments.

**Usage**

```
calcModfstat(exprs.dat, cl, class.vector)
```

**Arguments**

exprs.dat      a matrix of gene expression values.  
 cl             a vector of cluster assignments.  
 class.vector   a vector specifying group membership of the samples.

**Details**

This function is called internally by `findSynexprs`.

**Value**

a modified F-statistic (average MSS/average RSS) value for a set of cluster assignments.

**Author(s)**

Jessica Mar

**Examples**

```
## Not run:
library(cluster)
data(subset.loring.eset)
clustObj <- agnes(as.dist(1-t(cor(exprs(subset.loring.eset)))))
cfmod.vals <- NULL
for( i in 1:10 ){
cfmod.vals <- c(cfmod.vals, calcModfstat(exprs(subset.loring.eset), cutree(clustObj,i), pData(subset.loring.eset)
)
}
k <- (1:10)[cfmod.vals==max(cfmod.vals)]

## End(Not run)
```

---

calcRss

*Function calculates the average RSS for a set of cluster assignments.*

---

**Description**

Function calculates the average RSS for a set of cluster assignments.

**Usage**

```
calcRss(exprs.dat, cl, class.vector)
```

**Arguments**

`exprs.dat` a matrix of gene expression values.  
`cl` a vector of cluster assignments.  
`class.vector` a vector specifying the group membership of the samples.

**Details**

This function is called internally by `findSynexprs`. For an informative cluster, the RSS values should be very small relative to those produced by the informativeness metric (the MSS values).

**Value**

A numeric value representing the average RSS value for this set of cluster assignments.

**Author(s)**

Jessica Mar

**Examples**

```
## Not run:
library(cluster)
data(subset.loring.eset)
clustObj <- agnes(as.dist(1-t(cor(exprs(subset.loring.eset)))))
crss.vals <- NULL
for( i in 1:10 ){
  crss.vals <- c(crss.vals, calcRss(exprs(subset.loring.eset), cutree(clustObj,i), pData(subset.loring.eset)$cellt
)
}
# The RSS values are expected to be smaller than the informativeness metric values in the presence of genuine clusters

## End(Not run)
```

---

exprs.dat

*Gene Expression Matrix of Published Data*

---

**Description**

This is a matrix object containing published gene expression data from Mueller et al. (NCBI GEO accession id GSE11508). The data set contains 11044 probes for 68 samples. From the original data set, we have selected four cell lines giving a total of 68 samples - embryonic stem cells (12 samples), neural progenitors (31 samples), neural stem cells (8 samples) and teratoma-differentiated cells (17 samples). The lines have also been restricted based on Illumina BeadChip platform, and only those collected using the WG-6 version have been used.

We also applied a quality filter to the original gene expression data where a probe was retained if it passed a 0.99 detection score in 75

**Usage**

```
data(exprs.dat)
```

**Format**

A matrix with normalized log<sub>2</sub> expression intensities for 11044 probes on 68 samples (representing 4 different cell types).

**References**

Müller F, et al., Regulatory networks define phenotypic classes of human stem cell lines. Nature, 2008. 455(7211): p. 401-405.



**See Also**

[samp.info](#), [loring.eset](#)

**Examples**

```
data(exprs.dat)
```

---

findAttractors	<i>Infers the set of cell-lineage specific gene expression modules using GSEAlm and KEGG.</i>
----------------	---

---

**Description**

The function infers a set of KEGG pathways that correspond to the cell-lineage specific gene expression modules, as determined using GSEA. These pathways represent those that show the greatest discrimination between the different cell types or tissues in the expression data set supplied.

**Usage**

```
findAttractors(myEset, cellTypeTag, min.pwaysize = 5, annotation = "illuminaHumanv2BeadID.db", ...)
```

**Arguments**

myEset	ExpressionSet object.
cellTypeTag	character string of the variable name which stores the cell-lineages or experimental groups of interest for the samples in the data set (this string should be one of the column names of pData(myEset)).
min.pwaysize	integer specifying the minimum size of the KEGG pathways to consider in the analysis.
annotation	character string specifying the annotation package that corresponds to the chip platform the data was generated from.
...	additional arguments.

**Details**

This function subsets the expression data so that only those genes with annotations in KEGG are used for the downstream gene set enrichment analysis. This subset is stored in the eSet slot of the AttractoModuleSet output object.

The GSEAlm algorithm finds the KEGG pathway modules which discriminate between the cell-types or experimental groups of interest. It also ranks the results of the GSEAlm step by significance of these pathway modules, as stored in rankedPathways.

The output object of the findAttractors function also contains the incidence matrix that was built for the KEGG pathways, stored in the slot incidenceMatrix and the character string denoting which column of the sample data represents the cell type or experimental groups of interest, as stored in the slot cellTypeTag.

**Value**

An AttractorModuleSet object.

**Author(s)**

Jessica Mar

**References**

Jiang, Z. and R. Gentleman, Extensions to gene set enrichment. *Bioinformatics*, 2007. 23(3): p. 306-313. Kanehisa, M. and S. Goto, KEGG: Kyoto Encyclopedia of Genes and Genomes. *Nucleic Acids Res.*, 2000. 28: p. 27-30. Mar, J., C. Wells, and J. Quackenbush, Identifying the Gene Expression Modules that Represent the Drivers of Kauffman's Attractor Landscape. to appear, 2010.

**Examples**

```
data(subset.loring.eset)
attractor.states <- findAttractors(subset.loring.eset, "celltype", annotation="illuminaHumanv1.db")
```

---

findCorrPartners	<i>Determines Genes with Highly Correlated Expression Profiles to a Synexpression Group</i>
------------------	---

---

**Description**

This function finds genes with expression profiles highly correlated to a synexpression group.

**Usage**

```
findCorrPartners(mySynExpressionSet, myEset, removeGenes = NULL, cor.cutoff = 0.85, ...)
```

**Arguments**

mySynExpressionSet	SynExpressionSet object.
myEset	ExpressionSet object.
removeGenes	vector of probes that specify those genes who demonstrate little variability across the different celltypes and thus should be removed from downstream analysis.
cor.cutoff	numeric value specifying the correlation cut-off.
...	additional arguments.

**Details**

Genes with highly correlated profiles to the synexpression groups (e.g.  $R > 0.85$ ) are also likely to be integral in maintaining cell type-specific differences, however due to their lack of inclusion in resources like KEGG, would not have been picked up by the first GSEA step using findAttractors.

**Value**

A SynExpressionSet object which stores the genes that are highly correlated with the synexpression group provided, and their average expression profile.

**Author(s)**

Jessica Mar

**Examples**

```
data(subset.loring.eset)
attractor.states <- findAttractors(subset.loring.eset, "celltype", nperm=10, annotation="illuminaHumanv1.db")
remove.these.genes <- removeFlatGenes(subset.loring.eset, "celltype", contrasts=NULL, limma.cutoff=0.05)
mapk.syn <- findSynexprs("04010", attractor.states, remove.these.genes)
mapk.cor <- findCorrPartners(mapk.syn, subset.loring.eset, remove.these.genes)
```

---

findSynexprs	<i>This function finds the synexpression groups present within a core attractor pathway module.</i>
--------------	---

---

**Description**

This function takes the modules that were inferred from the GSEA step using (findAttractors) and finds a set of transcriptionally coherent set of genes associated with a particular core attractor pathway, i.e. the synexpression groups.

**Usage**

```
findSynexprs(pathwayIds, myAttractorModuleSet, removeGenes = NULL, min.clustersize = 5, ...)
```

**Arguments**

pathwayIds	either a single character string or vector of character strings denoting the KEGG IDs of the pathway modules to be analyzed.
myAttractorModuleSet	AttractorModuleSet object, output of the findAttractors step.
removeGenes	vector of gene names that specify those genes who demonstrate little variability across the different celltypes and thus should be removed from downstream analysis.
min.clustersize	integer specifying the minimum number of genes that must be present in clusters that are inferred.
...	additional arguments.

**Details**

This function performs a hierarchical cluster analysis of the genes in a core attractor pathway module, and uses an informativeness metric to determine the number of optimal clusters (synexpression groups) that describe the data.

**Value**

If a single KEGG ID is specified in `pwayIds`, then a `SynExpressionSet` object is returned. If a multiple KEGG IDs are specified, then an environment object is returned where the keys are labeled "pwayKEGGIDsynexprs" (e.g. for MAPK KEGGID = 04010, the key is `pway04010synexprs`). The value associated with each key is a `SynExpressionSet` object.

**Author(s)**

Jessica Mar

**References**

Mar, J., C. Wells, and J. Quackenbush, Identifying the Gene Expression Modules that Represent the Drivers of Kauffman's Attractor Landscape. to appear, 2010.

**Examples**

```
data(subset.loring.eset)
attractor.states <- findAttractors(subset.loring.eset, "celltype", nperm=10, annotation="illuminaHumanv1.db")
remove.these.genes <- removeFlatGenes(subset.loring.eset, "celltype", contrasts=NULL, limma.cutoff=0.05)
mapk.syn <- findSynexprs("04010", attractor.states, remove.these.genes)
top5.syn <- findSynexprs(attractor.states@rankedPathways[1:5,1], attractor.states, removeGenes=remove.these.genes)
```

---

loring.eset

NA

---

**Description**

This is an `ExpressionSet` object containing the published data from Müller et al. (NCBI GEO accession id GSE11508). The expression data set contains 11044 probes for 68 samples.

**Usage**

```
data(loring.eset)
```

**Format**

An `ExpressionSet` object.

**References**

Müller, F, et al., Regulatory networks define phenotypic classes of human stem cell lines. *Nature*, 2008. 455(7211): p. 401-405.

**See Also**

[exprs.dat](#), [samp.info](#)

**Examples**

```
data(loring.eset)
exprs.dat <- exprs(loring.eset) # gene expression matrix
```

---

plotsynexprs                      *Visualizing the Average Expression Profile of a Synexpression Group.*

---

**Description**

This function plots the average expression profile for a specific synexpression group.

**Usage**

```
plotsynexprs(mySynExpressionSet, tickMarks, tickLabels, vertLines, index=1, ...)
```

**Arguments**

mySynExpressionSet	<a href="#">SynExpressionSet</a> object.
tickMarks	numeric vector of specifying the location of the tick marks along the x-axis. There should be one tick for each cell type or group.
tickLabels	character vector specifying the labels to be appear underneath the tick marks on the x-axis. These should correspond to the cell type or group names.
vertLines	numeric vector specifying the location of the vertical lines that indicate the cell type or group-specific regions along the x-axis.
index	numeric value specifying which synexpression group should be plotted.
...	additional arguments.

**Details**

Generic plotting parameters can be passed to this function to create a more sophisticated plot, e.g `col="blue", main="Synexpression Group 1"`.

**Value**

A plot showing the average expression profile for the synexpression group specified.

**Author(s)**

Jessica Mar

**Examples**

```
## Not run:
data(subset.loring.eset)
attractor.states <- findAttractors(subset.loring.eset, "celltype", nperm=10, annotation="illuminaHumanv1.db")
remove.these.genes <- removeFlatGenes(subset.loring.eset, "celltype", contrasts=NULL, limma.cutoff=0.05)
mapk.syn <- findSynexprs("04010", attractor.states, remove.these.genes)
par(mfrow=c(2,2))
pretty.col <- rainbow(3)
for( i in 1:3 ){
plotsynexprs(mapk.syn, tickMarks=c(6, 28, 47, 60), tickLabels=c("ESC", "PRO", "NSC", "TER"), vertLines=c(12.5, 43.5, 60.5))
main=paste("Synexpression Group ", i, sep=""), col=pretty.col[i]
}

## End(Not run)
```

---

removeFlatGenes	<i>Flags a set of genes which demonstrates little variation across the celltypes or experimental groups of interest.</i>
-----------------	--

---

**Description**

This function uses a linear model set up in limma to assess the degree of association between celltype and a gene's expression profile. In this way, we can flag those genes whose profiles show very little change across different celltype groups, or in other words are "flat".

**Usage**

```
removeFlatGenes(eSet, cellTypeTag, contrasts = NULL, limma.cutoff = 0.05, ...)
```

**Arguments**

eSet	ExpressionSet object.
cellTypeTag	character string of the variable name which stores the cell-lineages or experimental groups of interest for the samples in the data set (this string should be one of the column names of pData(myEset)).
contrasts	optional vector of contrasts that specify the comparisons of interest. By default, all comparisons between the differnt groups are generated.
limma.cutoff	numeric specifying the P-value cutoff. Genes with P-values greater than this value are considered "flat" and will be included in the set of flat genes.
...	additional arguments.

**Details**

Flat genes are removed from the analysis after the core attractor pathway modules are first inferred (i.e. the findAttractors step).

**Value**

A vector with gene names (as defined in the eset) of those genes with expression profiles that hardly vary across different celltype or experimental groups.

**Author(s)**

Jessica Mar

**References**

limma package.

Smyth, G. K. (2004). Linear models and empirical Bayes methods for assessing differential expression in microarray experiments. *Statistical Applications in Genetics and Molecular Biology* 3, No. 1, Article 3.

**Examples**

```
data(subset.loring.eset)
remove.these.genes <- removeFlatGenes(subset.loring.eset, "celltype", contrasts=NULL, limma.cutoff=0.05)
```

---

somp.info

*somp.info* Contains the Sample Information for the Mueller data set.

---

**Description**

This is sample information data frame for the samples in the Mueller data set (NCBI GEO accession id GSE11508). The data frame contains the cell type groups for the 68 samples.

**Usage**

```
data(somp.info)
```

**Format**

A data.frame object with one column of sample IDs (these are the column IDs of the exprs.dat expression matrix object) and second column indicating which cell type each sample represents.

ChipID A vector of sample IDs.

celltype A vector denoting the cell type a sample represents.

**References**

Müller F, et al., Regulatory networks define phenotypic classes of human stem cell lines. *Nature*, 2008. 455(7211): p. 401-405.

**See Also**

[exprs.dat](#), [loring.eset](#)

## Examples

```
data(samp.info)
```

---

```
subset.loring.eset    NA
```

---

## Description

This is an ExpressionSet object containing a subset of the published data from Müller et al. (NCBI GEO accession id GSE11508). The expression data set contains 5522 probes for 68 samples. This ExpressionSet object was created specifically to demonstrate the functions in this package. If you're looking for the full Müller data set, see [loring.eset](#).

## Usage

```
data(subset.loring.eset)
```

## Format

An ExpressionSet object.

## References

Müller, F, et al., Regulatory networks define phenotypic classes of human stem cell lines. Nature, 2008. 455(7211): p. 401-405.

## See Also

[exprs.dat](#), [samp.info](#), [loring.eset](#)

## Examples

```
data(subset.loring.eset)
subset.exprs.dat <- exprs(subset.loring.eset) # gene expression matrix
```



---

SynExpressionSet-class

*Class SynExpressionSet*

---

### **Description**

This is a class representation for storing synexpression group information.

### **Objects from the Class**

Objects are output by the function [findSynexprs](#). Objects can also be created by using `new("SynExpressionSet", ...)`.

### **Slots**

**groups:** A list object denoting the probes belonging to each synexpression group.

**profiles:** A matrix of average expression profiles for each synexpression group, each profile is stored as a row.

### **Methods**

No methods have yet been defined with class "SynExpressionSet" in the signature.

### **Note**

This class is described in more detail in the vignette.

### **Author(s)**

Jessica Mar <jess@jimmy.harvard.edu>

### **Examples**

```
new.synexpressionset <- new("SynExpressionSet", groups=list(), profiles=matrix(0))
```

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