

Package ‘EnhancedVolcano’

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Type Package

Title Publication-ready volcano plots with enhanced colouring and labeling

Version 1.8.0

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Description Volcano plots represent a useful way to visualise the results of differential expression analyses. Here, we present a highly-configurable function that produces publication-ready volcano plots. EnhancedVolcano will attempt to fit as many point labels in the plot window as possible, thus avoiding 'clogging' up the plot with labels that could not otherwise have been read. Other functionality allows the user to identify up to 4 different types of attributes in the same plot space via colour, shape, size, and shade parameter configurations.

License GPL-3

Depends ggplot2, ggrepel

Imports ggalt, ggrastr

Suggests RUnit, BiocGenerics, knitr, DESeq2, pasilla, airway, org.Hs.eg.db, gridExtra, magrittr

URL <https://github.com/kevinblighe/EnhancedVolcano>

biocViews RNASeq, GeneExpression, Transcription, DifferentialExpression, ImmunoOncology

VignetteBuilder knitr

RoxygenNote 7.1.1

git_url <https://git.bioconductor.org/packages/EnhancedVolcano>

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R topics documented:

EnhancedVolcano 2

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Description

Volcano plots represent a useful way to visualise the results of differential expression analyses. Here, we present a highly-configurable function that produces publication-ready volcano plots [`@EnhancedVolcano`]. EnhancedVolcano will attempt to fit as many variable names in the plot window as possible, thus avoiding 'clogging' up the plot with labels that could not otherwise have been read.

Usage

```
EnhancedVolcano(
  toptable,
  lab,
  x,
  y,
  selectLab = NULL,
  xlim = c(min(toptable[[x]], na.rm = TRUE) - 1.5, max(toptable[[x]], na.rm = TRUE) +
    1.5),
  ylim = c(0, max(-log10(toptable[[y]]), na.rm = TRUE) + 5),
  xlab = bquote(~Log[2] ~ "fold change"),
  ylab = bquote(~-Log[10] ~ italic(P)),
  axisLabSize = 18,
  title = "Volcano plot",
  subtitle = bquote(italic(EnhancedVolcano)),
  caption = paste0("total = ", nrow(toptable), " variables"),
  titleLabSize = 18,
  subtitleLabSize = 14,
  captionLabSize = 14,
  pCutoff = 1e-05,
  FCcutoff = 1,
  cutoffLineType = "longdash",
  cutoffLineCol = "black",
  cutoffLineWidth = 0.4,
  pointSize = 2,
  labSize = 5,
  labCol = "black",
  labFace = "plain",
  labhjust = 0.5,
  labvjust = 1.5,
  boxedLabels = FALSE,
  shape = 19,
  shapeCustom = NULL,
```

```

col = c("grey30", "forestgreen", "royalblue", "red2"),
colCustom = NULL,
colAlpha = 1/2,
colGradient = NULL,
colGradientBreaks = c(pCutoff, 1),
colGradientLabels = c("0", "1.0"),
colGradientLimits = c(0, 1),
legendLabels = c("NS", expression(Log[2] ~ FC), "p-value", expression(p - value ~ and
  ~ log[2] ~ FC)),
legendPosition = "top",
legendLabSize = 14,
legendIconSize = 5,
legendDropLevels = TRUE,
encircle = NULL,
encircleCol = "black",
encircleFill = "pink",
encircleAlpha = 3/4,
encircleSize = 2.5,
shade = NULL,
shadeFill = "grey",
shadeAlpha = 1/2,
shadeSize = 0.01,
shadeBins = 2,
drawConnectors = FALSE,
widthConnectors = 0.5,
typeConnectors = "closed",
endsConnectors = "first",
lengthConnectors = unit(0.01, "npc"),
colConnectors = "grey10",
arrowheads = TRUE,
hline = NULL,
hlineType = "longdash",
hlineCol = "black",
hlineWidth = 0.4,
vline = NULL,
vlineType = "longdash",
vlineCol = "black",
vlineWidth = 0.4,
gridlines.major = TRUE,
gridlines.minor = TRUE,
border = "partial",
borderWidth = 0.8,
borderColour = "black",
raster = FALSE
)

```

Arguments

toptable A data-frame of test statistics (if not, a data frame, an attempt will be made to convert it to one). Requires at least the following: column for variable names (can be rownames); a column for log2 fold changes; a column for nominal or adjusted p-value.

lab	A column name in <code>toptable</code> containing variable names. Can be <code>rownames(toptable)</code> .
x	A column name in <code>toptable</code> containing \log_2 fold changes.
y	A column name in <code>toptable</code> containing nominal or adjusted p-values.
selectLab	A vector containing a subset of <code>lab</code> .
xlim	Limits of the x-axis.
ylim	Limits of the y-axis.
xlab	Label for x-axis.
ylab	Label for y-axis.
axisLabSize	Size of x- and y-axis labels.
title	Plot title.
subtitle	Plot subtitle.
caption	Plot caption.
titleLabSize	Size of plot title.
subtitleLabSize	Size of plot subtitle.
captionLabSize	Size of plot caption.
pCutoff	Cut-off for statistical significance. A horizontal line will be drawn at $-\log_{10}(\text{pCutoff})$.
FCcutoff	Cut-off for absolute \log_2 fold-change. Vertical lines will be drawn at the negative and positive values of $\log_2\text{FCcutoff}$.
cutoffLineType	Line type for <code>FCcutoff</code> and <code>pCutoff</code> ('blank', 'solid', 'dashed', 'dotted', 'dot-dash', 'longdash', 'twodash').
cutoffLineCol	Line colour for <code>FCcutoff</code> and <code>pCutoff</code> .
cutoffLineWidth	Line width for <code>FCcutoff</code> and <code>pCutoff</code> .
pointSize	Size of plotted points for each variable. Can be a single value or a vector of sizes.
labSize	Size of labels for each variable.
labCol	Colour of labels for each variable.
labFace	Font face of labels for each variable.
labhjust	Horizontal adjustment of label for each variable.
labvjust	Vertical adjustment of label for each variable.
boxedLabels	Logical, indicating whether or not to draw labels in boxes.
shape	Shape of the plotted points. Either a single value for all points, or 4 values corresponding to the default 4 legend labels specified by <code>legendLabels</code> .
shapeCustom	Named vector / key-value pairs that will over-ride the default shape scheme. The order must match that of <code>toptable</code> . Names / keys relate to groups / categories; values relate to shape encodings.
col	Colour shading for plotted points, corresponding to the default 4 legend labels specified by <code>legendLabels</code> .
colCustom	Named vector / key-value pairs that will over-ride the default colour scheme. The order must match that of <code>toptable</code> . Names / keys relate to groups / categories; values relate to colour.
colAlpha	Alpha for purposes of controlling colour transparency of variable points.

<code>colGradient</code>	If activated, over-rides the default discrete colour scheme and replaces it with a continuous scheme that shades based on nominal or adjusted p-value specified by <code>y</code> . For example, <code>c('red2', 'blue2')</code> .
<code>colGradientBreaks</code>	Break-points for the two colours specified by <code>colGradient</code> .
<code>colGradientLabels</code>	Labels for the break-points specified by <code>colGradientBreaks</code> .
<code>colGradientLimits</code>	Limits of the colour scheme specified by <code>colGradient</code> , i.e., max and min possible p-values.
<code>legendLabels</code>	Plot legend text labels.
<code>legendPosition</code>	Position of legend ('top', 'bottom', 'left', 'right').
<code>legendLabSize</code>	Size of plot legend text.
<code>legendIconSize</code>	Size of plot legend icons / symbols.
<code>legendDropLevels</code>	Logical, drop unused factor levels from legend.
<code>encircle</code>	A vector of variable names to encircle.
<code>encircleCol</code>	Colour of the encircled line.
<code>encircleFill</code>	Colour fill of the encircled region.
<code>encircleAlpha</code>	Alpha for purposes of controlling colour transparency of encircled region.
<code>encircleSize</code>	Line width of the encircled line.
<code>shade</code>	A vector of variable names to shade.
<code>shadeFill</code>	Colour of shaded regions.
<code>shadeAlpha</code>	Alpha for purposes of controlling colour transparency of shaded region.
<code>shadeSize</code>	Size of the shade contour lines.
<code>shadeBins</code>	Number of bins for the density of the shade.
<code>drawConnectors</code>	Logical, indicating whether or not to connect plot labels to their corresponding points by line connectors.
<code>widthConnectors</code>	Line width of connectors.
<code>typeConnectors</code>	Have the arrow head open ('open') or filled ('closed')?
<code>endsConnectors</code>	Which end of connectors to draw arrow head? ('last', 'first', 'both').
<code>lengthConnectors</code>	Length of the connectors.
<code>colConnectors</code>	Line colour of connectors.
<code>arrowheads</code>	Logical, indicating whether or not to draw arrow heads or or just have straight lines.
<code>hline</code>	Draw one or more horizontal lines passing through this/these values on y-axis. For single values, only a single numerical value is necessary. For multiple lines, pass these as a vector, e.g., <code>c(60,90)</code> .
<code>hlineType</code>	Line type for <code>hline</code> ('blank', 'solid', 'dashed', 'dotted', 'dotdash', 'longdash', 'twodash').
<code>hlineCol</code>	Colour of <code>hline</code> .
<code>hlineWidth</code>	Width of <code>hline</code> .

<code>vline</code>	Draw one or more vertical lines passing through this/these values on x-axis. For single values, only a single numerical value is necessary. For multiple lines, pass these as a vector, e.g., <code>c(60,90)</code> .
<code>vlineType</code>	Line type for <code>vline</code> ('blank', 'solid', 'dashed', 'dotted', 'dotdash', 'longdash', 'twodash').
<code>vlineCol</code>	Colour of <code>vline</code> .
<code>vlineWidth</code>	Width of <code>vline</code> .
<code>gridlines.major</code>	Logical, indicating whether or not to draw major gridlines.
<code>gridlines.minor</code>	Logical, indicating whether or not to draw minor gridlines.
<code>border</code>	Add a border for just the x and y axes ('partial') or the entire plot grid ('full')?
<code>borderWidth</code>	Width of the border on the x and y axes.
<code>borderColour</code>	Colour of the border on the x and y axes.
<code>raster</code>	Logical, indicating whether to rasterize the <code>geom_point</code> layer.

Details

Volcano plots represent a useful way to visualise the results of differential expression analyses. Here, we present a highly-configurable function that produces publication-ready volcano plots [[@EnhancedVolcano](#)]. `EnhancedVolcano` will attempt to fit as many variable names in the plot window as possible, thus avoiding 'clogging' up the plot with labels that could not otherwise have been read.

Value

A `ggplot2` object.

Author(s)

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Examples

```
library('pasilla')
pasCts <- system.file('extdata', 'pasilla_gene_counts.tsv',
  package='pasilla', mustWork=TRUE)
pasAnno <- system.file('extdata', 'pasilla_sample_annotation.csv',
  package='pasilla', mustWork=TRUE)
cts <- as.matrix(read.csv(pasCts, sep='\t', row.names='gene_id'))
coldata <- read.csv(pasAnno, row.names=1)
coldata <- coldata[,c('condition', 'type')]
rownames(coldata) <- sub('fb', '', rownames(coldata))
cts <- cts[, rownames(coldata)]
library('DESeq2')
dds <- DESeqDataSetFromMatrix(countData = cts,
  colData = coldata,
  design = ~ condition)

featureData <- data.frame(gene=rownames(cts))
mcols(dds) <- DataFrame(mcols(dds), featureData)
dds <- DESeq(dds)
```

```
res <- results(dds)

EnhancedVolcano(res,
  lab = rownames(res),
  x = 'log2FoldChange',
  y = 'pvalue',
  pCutoff = 10e-4,
  FCcutoff = 1.333,
  xlim = c(-5.5, 5.5),
  ylim = c(0, -log10(10e-12)),
  pointSize = 1.5,
  labSize = 2.5,
  title = 'DESeq2 results',
  subtitle = 'Differential expression',
  caption = 'FC cutoff, 1.333; p-value cutoff, 10e-4',
  legendPosition = "right",
  legendLabSize = 14,
  col = c('grey30', 'forestgreen', 'royalblue', 'red2'),
  colAlpha = 0.9,
  drawConnectors = TRUE,
  hline = c(10e-8),
  widthConnectors = 0.5)
```

Index

EnhancedVolcano, [2](#)

ggplot2, [6](#)