

Package ‘Maaslin2’

September 11, 2025

Title ``Multivariable Association Discovery in Population-scale
Meta-omics Studies"

Year 2021

Version 1.22.0

Depends R ($\geq$ 3.6)

Description MaAsLin2 is comprehensive R package for efficiently determining multivariable association between clinical metadata and microbial meta'omic features. MaAsLin2 relies on general linear models to accommodate most modern epidemiological study designs, including cross-sectional and longitudinal, and offers a variety of data exploration, normalization, and transformation methods. MaAsLin2 is the next generation of MaAsLin.

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LazyData false

Imports robustbase, biglm, pcaPP, edgeR, metagenomeSeq, pbapply, car,
dplyr, vegan, chemometrics, ggplot2, pheatmap, logging,
data.table, lmerTest, hash, optparse, grDevices, stats, utils,
glmmTMB, MASS, cplm, pscl, lme4, tibble

Suggests knitr, testthat ($\geq$ 2.1.0), rmarkdown, markdown

VignetteBuilder knitr

Collate fit.R utility_scripts.R viz.R Maaslin2.R

URL <http://huttenhower.sph.harvard.edu/maaslin2>

biocViews Metagenomics, Software, Microbiome, Normalization

BugReports <https://github.com/biobakery/maaslin2/issues>

git_url <https://git.bioconductor.org/packages/Maaslin2>

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Author Himel Mallick [aut],
 Ali Rahnavard [aut],
 Lauren McIver [aut, cre]

Maintainer Lauren McIver <lauren.j.mciver@gmail.com>

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Maaslin2	<i>MaAsLin2 is the next generation of MaAsLin, a multivariable statistical framework for finding associations between clinical metadata and potentially high-dimensional microbial multi-omics data.</i>
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Description

MaAsLin2 finds associations between microbiome meta-omics features and complex metadata in population-scale epidemiological studies. The software includes multiple analysis methods (including support for multiple covariates and repeated measures), filtering, normalization, and transform options to customize analysis for your specific study.

Usage

```
Maaslin2(
  input_data,
  input_metadata,
  output,
  min_abundance = 0.0,
  min_prevalence = 0.1,
  min_variance = 0.0,
  normalization = "TSS",
  transform = "LOG",
  analysis_method = "LM",
  max_significance = 0.25,
  random_effects = NULL,
  fixed_effects = NULL,
  correction = "BH",
  standardize = TRUE,
  cores = 1,
  plot_heatmap = TRUE,
  heatmap_first_n = 50,
  plot_scatter = TRUE,
  max_pngs = 10,
  save_scatter = FALSE,
  save_models = FALSE,
```

```

    reference = NULL
  )

```

Arguments

<code>input_data</code>	The tab-delimited input file of features.
<code>input_metadata</code>	The tab-delimited input file of metadata.
<code>output</code>	The output folder to write results.
<code>min_abundance</code>	The minimum abundance for each feature.
<code>min_prevalence</code>	The minimum percent of samples for which a feature is detected at minimum abundance.
<code>min_variance</code>	Keep features with variance greater than.
<code>max_significance</code>	The q-value threshold for significance.
<code>normalization</code>	The normalization method to apply.
<code>transform</code>	The transform to apply.
<code>analysis_method</code>	The analysis method to apply.
<code>random_effects</code>	The random effects for the model, comma-delimited for multiple effects.
<code>fixed_effects</code>	The fixed effects for the model, comma-delimited for multiple effects.
<code>correction</code>	The correction method for computing the q-value.
<code>standardize</code>	Apply z-score so continuous metadata are on the same scale.
<code>plot_heatmap</code>	Generate a heatmap for the significant associations.
<code>heatmap_first_n</code>	In heatmap, plot top N features with significant associations.
<code>plot_scatter</code>	Generate scatter plots for the significant associations.
<code>max_pngs</code>	Set the maximum number of scatter plots for significant associations to save as png files.
<code>save_scatter</code>	Save all scatter plot ggplot objects to an RData file.
<code>cores</code>	The number of R processes to run in parallel.
<code>save_models</code>	Return the full model outputs and save to an RData file.
<code>reference</code>	The factor to use as a reference for a variable with more than two levels provided as a string of 'variable,reference' semi-colon delimited for multiple variables.

Value

List containing the results from applying the model.

Author(s)

Himel Mallick<himel.stat.iitk@gmail.com>,
 Ali Rahnavard<gholamali.rahnavaard@gmail.com>,
 Maintainers: Lauren McIver<lauren.j.mciver@gmail.com>,

Examples

```
input_data <- system.file(
  'extdata', 'HMP2_taxonomy.tsv', package="Maaslin2")
input_metadata <- system.file(
  'extdata', 'HMP2_metadata.tsv', package="Maaslin2")
fit_data <- Maaslin2(
  input_data, input_metadata, 'demo_output', transform = "AST",
  fixed_effects = c('diagnosis', 'dysbiosisnonIBD', 'dysbiosisUC', 'dysbiosisCD', 'antibiotics', 'age'),
  random_effects = c('site', 'subject'),
  normalization = 'NONE',
  reference = 'diagnosis,nonIBD',
  standardize = FALSE)
```

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