

Package ‘CRMetrics’

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Title Cell Ranger Output Filtering and Metrics Visualization

Version 0.3.2

Description Sample and cell filtering as well as visualisation of output metrics from 'Cell Ranger' by Grace X.Y. Zheng et al. (2017) <[doi:10.1038/ncomms14049](https://doi.org/10.1038/ncomms14049)>. 'CRMetrics' allows for easy plotting of output metrics across multiple samples as well as comparative plots including statistical assessments of these. 'CRMetrics' allows for easy removal of ambient RNA using 'SoupX' by Matthew D Young and Sam Behjati (2020) <[doi:10.1093/gigascience/giaa151](https://doi.org/10.1093/gigascience/giaa151)> or 'CellBender' by Stephen J Fleming et al. (2022) <[doi:10.1101/791699](https://doi.org/10.1101/791699)>. Furthermore, it is possible to preprocess data using 'Pagoda2' by Nikolas Barkas et al. (2021) <<https://github.com/kharchenkolab/pagoda2>> or 'Seurat' by Yuhan Hao et al. (2021) <[doi:10.1016/j.cell.2021.04.048](https://doi.org/10.1016/j.cell.2021.04.048)> followed by embedding of cells using 'Conos' by Nikolas Barkas et al. (2019) <[doi:10.1038/s41592-019-0466-z](https://doi.org/10.1038/s41592-019-0466-z)>. Finally, doublets can be detected using 'scrublet' by Samuel L. Wolock et al. (2019) <[doi:10.1016/j.cels.2018.11.005](https://doi.org/10.1016/j.cels.2018.11.005)> or 'Doublet Detection' by Gayoso et al. (2020) <[doi:10.5281/zenodo.2678041](https://doi.org/10.5281/zenodo.2678041)>. In the end, cells are filtered based on user input for use in downstream applications.

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Depends R (>= 4.0.0)

Imports cowplot, dplyr, ggbeeswarm, ggplot2, ggpmisc, ggpubr, ggrepel, magrittr, Matrix, methods, R6, scales, sccore, sparseMatrixStats, stats, tibble, tidyr, utils

Suggests conos, data.table, markdown, pagoda2, reticulate, rhdf5, Seurat, SoupX, testthat (>= 3.0.0)

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URL <https://github.com/khodosevichlab/CRMetrics>

BugReports <https://github.com/khodosevichlab/CRMetrics/issues>

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Description

Functions to analyze Cell Ranger count data. To initialize a new object, 'data.path' or 'cms' is needed. 'metadata' is also recommended, but not required.

Public fields

`metadata` data.frame or character Path to metadata file or name of metadata data.frame object. Metadata must contain a column named 'sample' containing sample names that must match folder names in 'data.path' (default = NULL)

`data.path` character Path(s) to Cell Ranger count data, one directory per sample. If multiple paths, do c("path1", "path2") (default = NULL)

`cms` list List with count matrices (default = NULL)

`cms.preprocessed` list List with preprocessed count matrices after `$doPreprocessing()` (default = NULL)

`cms.raw` list List with raw, unfiltered count matrices, i.e., including all CBs detected also empty droplets (default = NULL)

`summary.metrics` data.frame Summary metrics from Cell Ranger (default = NULL)

`detailed.metrics` data.frame Detailed metrics, i.e., no. genes and UMIs per cell (default = NULL)

`comp.group` character A group present in the metadata to compare the metrics by, can be added with `addComparison` (default = NULL)

`verbose` logical Print messages or not (default = TRUE)

`theme` ggplot2 theme (default: `theme_bw()`)

`pal` Plotting palette (default = NULL)

`n.cores` numeric Number of cores for calculations (default = 1) Initialize a `CRMetrics` object

Methods

Public methods:

- `CRMetrics$new()`
- `CRMetrics$addDetailedMetrics()`
- `CRMetrics$addComparison()`
- `CRMetrics$plotSamples()`
- `CRMetrics$plotSummaryMetrics()`
- `CRMetrics$plotDetailedMetrics()`
- `CRMetrics$plotEmbedding()`
- `CRMetrics$plotDepth()`
- `CRMetrics$plotMitoFraction()`
- `CRMetrics$detectDoublets()`
- `CRMetrics$doPreprocessing()`
- `CRMetrics$createEmbedding()`
- `CRMetrics$filterCms()`
- `CRMetrics$selectMetrics()`
- `CRMetrics$plotFilteredCells()`
- `CRMetrics$getDepth()`
- `CRMetrics$getMitoFraction()`
- `CRMetrics$prepareCellbender()`
- `CRMetrics$saveCellbenderScript()`
- `CRMetrics$getExpectedCells()`
- `CRMetrics$getTotalDroplets()`
- `CRMetrics$addCms()`
- `CRMetrics$plotCbTraining()`
- `CRMetrics$plotCbCellProbs()`
- `CRMetrics$plotCbAmbExp()`
- `CRMetrics$plotCbAmbGenes()`
- `CRMetrics$addSummaryFromCms()`
- `CRMetrics$runSoupX()`
- `CRMetrics$plotSoupX()`
- `CRMetrics$plotCbCells()`
- `CRMetrics$addDoublets()`
- `CRMetrics$clone()`

Method `new()`: To initialize new object, 'data.path' or 'cms' is needed. 'metadata' is also recommended, but not required.

Usage:

```
CRMetrics$new(  
  data.path = NULL,  
  metadata = NULL,  
  cms = NULL,  
  samples = NULL,
```

```

unique.names = TRUE,
sep.cells = "!!",
comp.group = NULL,
verbose = TRUE,
theme = theme_bw(),
n.cores = 1,
sep.meta = ",",
raw.meta = FALSE,
pal = NULL
)

```

Arguments:

`data.path` character Path to directory with Cell Ranger count data, one directory per sample (default = NULL).

`metadata` data.frame or character Path to metadata file (comma-separated) or name of metadata dataframe object. Metadata must contain a column named 'sample' containing sample names that must match folder names in 'data.path' (default = NULL)

`cms` list List with count matrices (default = NULL)

`samples` character Sample names. Only relevant if cms is provided (default = NULL)

`unique.names` logical Create unique cell names. Only relevant if cms is provided (default = TRUE)

`sep.cells` character Sample-cell separator. Only relevant if cms is provided and unique.names=TRUE (default = "!!")

`comp.group` character A group present in the metadata to compare the metrics by, can be added with addComparison (default = NULL)

`verbose` logical Print messages or not (default = TRUE)

`theme` ggplot2 theme (default: theme_bw())

`n.cores` integer Number of cores for the calculations (default = self\$n.cores)

`sep.meta` character Separator for metadata file (default = ",")

`raw.meta` logical Keep metadata in its raw format. If FALSE, classes will be converted using "type.convert" (default = FALSE)

`pal` character Plotting palette (default = NULL)

Returns: CRMetrics object

Examples:

```

\dontrun{
crm <- CRMetrics$new(data.path = "/path/to/count/data/")
}

```

Method addDetailedMetrics(): Function to read in detailed metrics. This is not done upon initialization for speed.

Usage:

```

CRMetrics$new()$addDetailedMetrics(
  cms = self$cms,
  min.transcripts.per.cell = 100,
  n.cores = self$n.cores,
  verbose = self$verbose
)

```

Arguments:

cms list List of (sparse) count matrices (default = self\$cms)
 min.transcripts.per.cell numeric Minimal number of transcripts per cell (default = 100)
 n.cores integer Number of cores for the calculations (default = self\$n.cores).
 verbose logical Print messages or not (default = self\$verbose).

Returns: Count matrices

Examples:

```
# Simulate data
testdata.cms <- lapply(seq_len(2), \(x) {
  out <- Matrix::rsparsematrix(2e3, 1e3, 0.1)
  out[out < 0] <- 1
  dimnames(out) <- list(sapply(seq_len(2e3), \(x) paste0("gene",x)),
    sapply(seq_len(1e3), \(x) paste0("cell",x)))
  return(out)
})

# Initialize
crm <- CRMetrics$new(cms = testdata.cms, samples = c("sample1", "sample2"), n.cores = 1)

# Run function
crm$addDetailedMetrics()
```

Method addComparison(): Add comparison group for statistical testing.

Usage:

```
CRMetrics$addComparison(comp.group, metadata = self$metadata)
```

Arguments:

comp.group character Comparison metric (default = self\$comp.group).
 metadata data.frame Metadata for samples (default = self\$metadata).

Returns: Vector

Examples:

```
# Simulate data
testdata.cms <- lapply(seq_len(2), \(x) {
  out <- Matrix::rsparsematrix(2e3, 1e3, 0.1)
  out[out < 0] <- 1
  dimnames(out) <- list(sapply(seq_len(2e3), \(x) paste0("gene",x)),
    sapply(seq_len(1e3), \(x) paste0("cell",x)))
  return(out)
})

# Initialize
crm <- CRMetrics$new(cms = testdata.cms, samples = c("sample1", "sample2"), n.cores = 1)

# Add metadata
crm$metadata <- data.frame(sex = c("male", "female"))
```

```
# Add comparison group
crm$addComparison(comp.group = "sex")
```

Method `plotSamples()`: Plot the number of samples.

Usage:

```
CRMetrics$plotSamples(
  comp.group = self$comp.group,
  h.adj = 0.05,
  exact = FALSE,
  metadata = self$metadata,
  second.comp.group = NULL,
  pal = self$pal
)
```

Arguments:

`comp.group` character Comparison metric, must match a column name of metadata (default = `self$comp.group`).

`h.adj` numeric Position of statistics test p value as % of max(y) (default = 0.05).

`exact` logical Whether to calculate exact p values (default = FALSE).

`metadata` data.frame Metadata for samples (default = `self$metadata`).

`second.comp.group` character Second comparison metric, must match a column name of metadata (default = NULL).

`pal` character Plotting palette (default = `self$pal`)

Returns: ggplot2 object

Examples:

```
samples <- c("sample1", "sample2")

# Simulate data
testdata.cms <- lapply(seq_len(2), \x) {
  out <- Matrix::rsparsematrix(2e3, 1e3, 0.1)
  out[out < 0] <- 1
  dimnames(out) <- list(sapply(seq_len(2e3), \x) paste0("gene",x)),
    sapply(seq_len(1e3), \x) paste0("cell",x))
  return(out)
})
names(testdata.cms) <- samples

# Create metadata
metadata <- data.frame(sample = samples,
  sex = c("male", "female"),
  condition = c("a", "b"))

# Initialize
crm <- CRMetrics$new(cms = testdata.cms, metadata = metadata, n.cores = 1)

# Plot
crm$plotSamples(comp.group = "sex", second.comp.group = "condition")
```

Method plotSummaryMetrics(): Plot all summary stats or a selected list.

Usage:

```
CRMetrics$plotSummaryMetrics(
  comp.group = self$comp.group,
  second.comp.group = NULL,
  metrics = NULL,
  h.adj = 0.05,
  plot.stat = TRUE,
  stat.test = c("non-parametric", "parametric"),
  exact = FALSE,
  metadata = self$metadata,
  summary.metrics = self$summary.metrics,
  plot.geom = "bar",
  se = FALSE,
  group.reg.lines = FALSE,
  secondary.testing = TRUE,
  pal = self$pal
)
```

Arguments:

`comp.group` character Comparison metric (default = `self$comp.group`).

`second.comp.group` character Second comparison metric, used for the metric "samples per group" or when "comp.group" is a numeric or an integer (default = `NULL`).

`metrics` character Metrics to plot (default = `NULL`).

`h.adj` numeric Position of statistics test p value as % of max(y) (default = 0.05)

`plot.stat` logical Show statistics in plot. Will be `FALSE` if "comp.group" = "sample" or if "comp.group" is a numeric or an integer (default = `TRUE`)

`stat.test` character Statistical test to perform to compare means. Can either be "non-parametric" or "parametric" (default = "non-parametric").

`exact` logical Whether to calculate exact p values (default = `FALSE`).

`metadata` data.frame Metadata for samples (default = `self$metadata`).

`summary.metrics` data.frame Summary metrics (default = `self$summary.metrics`).

`plot.geom` character Which geometric is used to plot the data (default = "point").

`se` logical For regression lines, show SE (default = `FALSE`)

`group.reg.lines` logical For regression lines, if `FALSE` show one line, if `TRUE` show line per group defined by `second.comp.group` (default = `FALSE`)

`secondary.testing` logical Whether to show post hoc testing (default = `TRUE`)

`pal` character Plotting palette (default = `self$pal`)

Returns: ggplot2 object

Examples:

```
\donttest{
# Simulate data
testdata.cms <- lapply(seq_len(2), \(x) {
  out <- Matrix::rsparsematrix(2e3, 1e3, 0.1)
  out[out < 0] <- 1
  dimnames(out) <- list(sapply(seq_len(2e3), \(x) paste0("gene", x)),
```

```

sapply(seq_len(1e3), \(x) paste0("cell",x)))
return(out)
})

# Initialize
crm <- CRMetrics$new(cms = testdata.cms, samples = c("sample1", "sample2"), n.cores = 1)

# Add summary metrics
crm$addSummaryFromCms()

crm$plotSummaryMetrics(plot.geom = "point")
}

```

Method `plotDetailedMetrics()`: Plot detailed metrics from the `detailed.metrics` object

Usage:

```

CRMetrics$plotDetailedMetrics(
  comp.group = self$comp.group,
  detailed.metrics = self$detailed.metrics,
  metadata = self$metadata,
  metrics = NULL,
  plot.geom = "violin",
  hline = TRUE,
  pal = self$pal
)

```

Arguments:

`comp.group` character Comparison metric (default = `self$comp.group`).

`detailed.metrics` data.frame Object containing the count matrices (default = `self$detailed.metrics`).

`metadata` data.frame Metadata for samples (default = `self$metadata`).

`metrics` character Metrics to plot. NULL plots both plots (default = NULL).

`plot.geom` character How to plot the data (default = "violin").

`hline` logical Whether to show median as horizontal line (default = TRUE)

`pal` character Plotting palette (default = `self$pal`)

`data.path` character Path to Cell Ranger count data (default = `self$data.path`).

Returns: ggplot2 object

Examples:

```

\donttest{
# Simulate data
testdata.cms <- lapply(seq_len(2), \(x) {
out <- Matrix::rsparsematrix(2e3, 1e3, 0.1)
out[out < 0] <- 1
dimnames(out) <- list(sapply(seq_len(2e3), \(x) paste0("gene",x)),
sapply(seq_len(1e3), \(x) paste0("cell",x)))
return(out)
})

# Initialize

```



```

crm <- CRMetrics$new(cms = testdata.cms, samples = c("sample1", "sample2"), n.cores = 1)

# Add detailed metrics
crm$addDetailedMetrics()

# Plot
crm$plotDetailedMetrics()
}

```

Method `plotEmbedding()`: Plot cells in embedding using Conos and color by depth and doublets.

Usage:

```

CRMetrics$plotEmbedding(
  depth = FALSE,
  doublet.method = NULL,
  doublet.scores = FALSE,
  depth.cutoff = 1000,
  mito.frac = FALSE,
  mito.cutoff = 0.05,
  species = c("human", "mouse"),
  size = 0.3,
  sep = "!!",
  pal = NULL,
  ...
)

```

Arguments:

`depth` logical Plot depth or not (default = FALSE).
`doublet.method` character Doublet detection method (default = NULL).
`doublet.scores` logical Plot doublet scores or not (default = FALSE).
`depth.cutoff` numeric Depth cutoff (default = 1e3).
`mito.frac` logical Plot mitochondrial fraction or not (default = FALSE).
`mito.cutoff` numeric Mitochondrial fraction cutoff (default = 0.05).
`species` character Species to calculate the mitochondrial fraction for (default = c("human", "mouse")).
`size` numeric Dot size (default = 0.3)
`sep` character Separator for creating unique cell names (default = "!!")
`pal` character Plotting palette (default = NULL)
... Plotting parameters passed to `score::embeddingPlot`.

Returns: ggplot2 object

Examples:

```

\donttest{
if (requireNamespace("pagoda2", quietly = TRUE)) {
if (requireNamespace("conos", quietly = TRUE)) {
# Simulate data
testdata.cms <- lapply(seq_len(2), \(x) {
out <- Matrix::rsparsematrix(2e3, 1e3, 0.1)

```

```

out[out < 0] <- 1
dimnames(out) <- list(sapply(seq_len(2e3), \(x) paste0("gene",x)),
sapply(seq_len(1e3), \(x) paste0("cell",x)))
return(out)
})

# Initialize
crm <- CRMetrics$new(cms = testdata.cms, samples = c("sample1", "sample2"), n.cores = 1)

# Create embedding
crm$doPreprocessing()
crm$createEmbedding()

crm$plotEmbedding()
} else {
message("Package 'conos' not available.")
}
} else {
message("Package 'pagoda2' not available.")
}
}
}

```

Method `plotDepth()`: Plot the sequencing depth in histogram.

Usage:

```

CRMetrics$plotDepth(
  cutoff = 1000,
  samples = self$metadata$sample,
  sep = "!!",
  keep.col = "#E7CDC2",
  filter.col = "#A65141"
)

```

Arguments:

`cutoff` numeric The depth cutoff to color the cells in the embedding (default = 1e3).
`samples` character Sample names to include for plotting (default = `$metadata$sample`).
`sep` character Separator for creating unique cell names (default = "!!")
`keep.col` character Color for density of cells that are kept (default = "#E7CDC2")
`filter.col` Character Color for density of cells to be filtered (default = "#A65141")

Returns: ggplot2 object

Examples:

```

\donttest{
if (requireNamespace("pagoda2", quietly = TRUE)) {
if (requireNamespace("conos", quietly = TRUE)) {
# Simulate data
testdata.cms <- lapply(seq_len(2), \(x) {
out <- Matrix::rsparsematrix(2e3, 1e3, 0.1)
out[out < 0] <- 1

```

```

dimnames(out) <- list(sapply(seq_len(2e3), \(x) paste0("gene",x)),
sapply(seq_len(1e3), \(x) paste0("cell",x)))
return(out)
})

# Initialize
crm <- CRMetrics$new(cms = testdata.cms, samples = c("sample1", "sample2"), n.cores = 1)

# Create embedding
crm$doPreprocessing()
crm$createEmbedding()

# Plot
crm$plotDepth()
} else {
message("Package 'conos' not available.")
}
} else {
message("Package 'pagoda2' not available.")
}
}
}

```

Method `plotMitoFraction()`: Plot the mitochondrial fraction in histogram.

Usage:

```

CRMetrics$plotMitoFraction(
  cutoff = 0.05,
  species = c("human", "mouse"),
  samples = self$metadata$sample,
  sep = "!!!",
  keep.col = "#E7CDC2",
  filter.col = "#A65141"
)

```

Arguments:

`cutoff` numeric The mito. fraction cutoff to color the embedding (default = 0.05)
`species` character Species to calculate the mitochondrial fraction for (default = "human")
`samples` character Sample names to include for plotting (default = `$metadata$sample`)
`sep` character Separator for creating unique cell names (default = "!!!")
`keep.col` character Color for density of cells that are kept (default = "#E7CDC2")
`filter.col` Character Color for density of cells to be filtered (default = "#A65141")

Returns: ggplot2 object

Examples:

```

\donttest{
if (requireNamespace("pagoda2", quietly = TRUE)) {
if (requireNamespace("conos", quietly = TRUE)) {
# Simulate data
testdata.cms <- lapply(seq_len(2), \(x) {

```

```

out <- Matrix::rsparsematrix(2e3, 1e3, 0.1)
out[out < 0] <- 1
dimnames(out) <- list(sapply(seq_len(2e3), \(x) paste0("gene",x)),
sapply(seq_len(1e3), \(x) paste0("cell",x)))
return(out)
})

# Initialize
crm <- CRMetrics$new(cms = testdata.cms, samples = c("sample1", "sample2"), n.cores = 1)

# Create embedding
crm$doPreprocessing()
crm$createEmbedding()

# Plot
crm$plotMitoFraction()
} else {
message("Package 'conos' not available.")
}
} else {
message("Package 'pagoda2' not available.")
}
}
}

```

Method detectDoublets(): Detect doublet cells.

Usage:

```

CRMetrics$detectDoublets(
  method = c("scrublet", "doubletdetection"),
  cms = self$cms,
  samples = self$metadata$sample,
  env = "r-reticulate",
  conda.path = system("whereis conda"),
  n.cores = self$n.cores,
  verbose = self$verbose,
  args = list(),
  export = FALSE,
  data.path = self$data.path
)

```

Arguments:

method character Which method to use, either scrublet or doubletdetection (default="scrublet").

cms list List containing the count matrices (default=self\$cms).

samples character Vector of sample names. If NULL, samples are extracted from cms (default = self\$metadata\$sample)

env character Environment to run python in (default="r-reticulate").

conda.path character Path to conda environment (default=system("whereis conda")).

n.cores integer Number of cores to use (default = self\$n.cores)

verbose logical Print messages or not (default = self\$verbose)

`args` list A list with additional arguments for either `DoubletDetection` or `scrublet`. Please check the respective manuals.

`export` boolean Export CMs in order to detect doublets outside R (default = FALSE)

`data.path` character Path to write data, only relevant if `export = TRUE`. Last character must be `/` (default = `self$data.path`)

Returns: data.frame

Examples:

```
\dontrun{
# Simulate data
testdata.cms <- lapply(seq_len(2), \(x) {
out <- Matrix::rsparsematrix(2e3, 1e3, 0.1)
out[out < 0] <- 1
dimnames(out) <- list(sapply(seq_len(2e3), \(x) paste0("gene",x)),
sapply(seq_len(1e3), \(x) paste0("cell",x)))
return(out)
})

# Initialize
crm <- CRMetrics$new(cms = testdata.cms, samples = c("sample1", "sample2"), n.cores = 1)

# Detect doublets
crm$detectDoublets(method = "scrublet",
conda.path = "/opt/software/miniconda/4.12.0/condabin/conda")
}
```

Method `doPreprocessing()`: Perform conos preprocessing.

Usage:

```
CRMetrics$doPreprocessing(
  cms = self$cms,
  preprocess = c("pagoda2", "seurat"),
  min.transcripts.per.cell = 100,
  verbose = self$verbose,
  n.cores = self$n.cores,
  get.largevis = FALSE,
  tsne = FALSE,
  make.geneknn = FALSE,
  cluster = FALSE,
  ...
)
```

Arguments:

`cms` list List containing the count matrices (default = `self$cms`).

`preprocess` character Method to use for preprocessing (default = `c("pagoda2","seurat")`).

`min.transcripts.per.cell` numeric Minimal transcripts per cell (default = 100)

`verbose` logical Print messages or not (default = `self$verbose`).

`n.cores` integer Number of cores for the calculations (default = `self$n.cores`).

```

get.largevis logical For Pagoda2, create largeVis embedding (default = FALSE)
tsne logical Create tSNE embedding (default = FALSE)
make.geneknn logical For Pagoda2, estimate gene kNN (default = FALSE)
cluster logical For Seurat, estimate clusters (default = FALSE)
... Additional arguments for Pagaoda2::basicP2Proc or conos::basicSeuratProc

```

Returns: Conos object

Examples:

```

\donttest{
if (requireNamespace("pagoda2", quietly = TRUE)) {
# Simulate data
testdata.cms <- lapply(seq_len(2), \(x) {
out <- Matrix::rsparsematrix(2e3, 1e3, 0.1)
out[out < 0] <- 1
dimnames(out) <- list(sapply(seq_len(2e3), \(x) paste0("gene",x)),
sapply(seq_len(1e3), \(x) paste0("cell",x)))
return(out)
})

# Initialize
crm <- CRMetrics$new(cms = testdata.cms, samples = c("sample1", "sample2"), n.cores = 1)

# Perform preprocessing
crm$doPreprocessing(preprocess = "pagoda2")
} else {
message("Package 'pagoda2' not available.")
}
}

```

Method createEmbedding(): Create Conos embedding.

Usage:

```

CRMetrics$createEmbedding(
  cms = self$cms.preprocessed,
  verbose = self$verbose,
  n.cores = self$n.cores,
  arg.buildGraph = list(),
  arg.findCommunities = list(),
  arg.embedGraph = list(method = "UMAP")
)

```

Arguments:

```

cms list List containing the preprocessed count matrices (default = self$cms.preprocessed).
verbose logical Print messages or not (default = self$verbose).
n.cores integer Number of cores for the calculations (default = self$n.cores).
arg.buildGraph list A list with additional arguments for the buildGraph function in Conos
(default = list())
arg.findCommunities list A list with additional arguments for the findCommunities function
in Conos (default = list())

```

`arg.embedGraph` list A list with additional arguments for the `embedGraph` function in `Conos` (default = `list(method = "UMAP")`)

Returns: `Conos` object

Examples:

```
\donttest{
if (requireNamespace("pagoda2", quietly = TRUE)) {
if (requireNamespace("conos", quietly = TRUE)) {
# Simulate data
testdata.cms <- lapply(seq_len(2), \(\x) {
out <- Matrix::rsparsematrix(2e3, 1e3, 0.1)
out[out < 0] <- 1
dimnames(out) <- list(sapply(seq_len(2e3), \(\x) paste0("gene",x)),
sapply(seq_len(1e3), \(\x) paste0("cell",x)))
return(out)
})

# Initialize
crm <- CRMetrics$new(cms = testdata.cms, samples = c("sample1", "sample2"), n.cores = 1)

# Create embedding
crm$doPreprocessing()
crm$createEmbedding()
} else {
message("Package 'conos' not available.")
}
} else {
message("Package 'pagoda2' not available.")
}
}
```

Method `filterCms()`: Filter cells based on depth, mitochondrial fraction and doublets from the count matrix.

Usage:

```
CRMetrics$filterCms(
  depth.cutoff = NULL,
  mito.cutoff = NULL,
  doublets = NULL,
  species = c("human", "mouse"),
  samples.to.exclude = NULL,
  verbose = self$verbose,
  sep = "!!",
  raw = FALSE
)
```

Arguments:

`depth.cutoff` numeric Depth (transcripts per cell) cutoff (default = `NULL`).

`mito.cutoff` numeric Mitochondrial fraction cutoff (default = `NULL`).

`doublets` character Doublet detection method to use (default = `NULL`).

species character Species to calculate the mitochondrial fraction for (default = "human").
 samples.to.exclude character Sample names to exclude (default = NULL)
 verbose logical Show progress (default = self\$verbose)
 sep character Separator for creating unique cell names (default = "!!")
 raw boolean Filter on raw, unfiltered count matrices. Usually not intended (default = FALSE)

Returns: list of filtered count matrices

Examples:

```
\donttest{
if (requireNamespace("pagoda2", quietly = TRUE)) {
if (requireNamespace("conos", quietly = TRUE)) {
# Simulate data
testdata.cms <- lapply(seq_len(2), \(x) {
out <- Matrix::rsparsematrix(2e3, 1e3, 0.1)
out[out < 0] <- 1
dimnames(out) <- list(sapply(seq_len(2e3), \(x) paste0("gene",x)),
sapply(seq_len(1e3), \(x) paste0("cell",x)))
return(out)
})

# Initialize
crm <- CRMetrics$new(cms = testdata.cms, samples = c("sample1", "sample2"), n.cores = 1)

# Create embedding
crm$doPreprocessing()
crm$createEmbedding()

# Filter CMs
crm$filterCms(depth.cutoff = 1e3, mito.cutoff = 0.05)
} else {
message("Package 'conos' not available.")
}
} else {
message("Package 'pagoda2' not available.")
}
}
```

Method selectMetrics(): Select metrics from summary.metrics

Usage:

```
CRMetrics$selectMetrics(ids = NULL)
```

Arguments:

ids character Metric id to select (default = NULL).

Returns: vector

Examples:


```

# Simulate data
testdata.cms <- lapply(seq_len(2), \(x) {
  out <- Matrix::rsparsematrix(2e3, 1e3, 0.1)
  out[out < 0] <- 1
  dimnames(out) <- list(sapply(seq_len(2e3), \(x) paste0("gene",x)),
    sapply(seq_len(1e3), \(x) paste0("cell",x)))
  return(out)
})

# Initialize
crm <- CRMetrics$new(cms = testdata.cms, samples = c("sample1", "sample2"), n.cores = 1)

# Select metrics
crm$selectMetrics()
selection.metrics <- crm$selectMetrics(c(1:4))

```

Method `plotFilteredCells()`: Plot filtered cells in an embedding, in a bar plot, on a tile or export the data frame

Usage:

```

CRMetrics$plotFilteredCells(
  type = c("embedding", "bar", "tile", "export"),
  depth = TRUE,
  depth.cutoff = 1000,
  doublet.method = NULL,
  mito.frac = TRUE,
  mito.cutoff = 0.05,
  species = c("human", "mouse"),
  size = 0.3,
  sep = "!!",
  cols = c("grey80", "red", "blue", "green", "yellow", "black", "pink", "purple"),
  ...
)

```

Arguments:

`type` character The type of plot to use: embedding, bar, tile or export (default = c("embedding","bar","tile","export")).

`depth` logical Plot the depth or not (default = TRUE).

`depth.cutoff` numeric Depth cutoff, either a single number or a vector with cutoff per sample and with sampleIDs as names (default = 1e3).

`doublet.method` character Method to detect doublets (default = NULL).

`mito.frac` logical Plot the mitochondrial fraction or not (default = TRUE).

`mito.cutoff` numeric Mitochondrial fraction cutoff, either a single number or a vector with cutoff per sample and with sampleIDs as names (default = 0.05).

`species` character Species to calculate the mitochondrial fraction for (default = c("human","mouse")).

`size` numeric Dot size (default = 0.3)

`sep` character Separator for creating unique cell names (default = "!!")

`cols` character Colors used for plotting (default = c("grey80","red","blue","green","yellow","black","pink","purple"))

... Plotting parameters passed to `score::embeddingPlot`.

Returns: ggplot2 object or data frame

Examples:

```
\donttest{
if (requireNamespace("pagoda2", quietly = TRUE)) {
if (requireNamespace("conos", quietly = TRUE)) {
# Simulate data
testdata.cms <- lapply(seq_len(2), \x) {
out <- Matrix::rsparsematrix(2e3, 1e3, 0.1)
out[out < 0] <- 1
dimnames(out) <- list(sapply(seq_len(2e3), \x) paste0("gene",x)),
sapply(seq_len(1e3), \x) paste0("cell",x))
return(out)
})

# Initialize
crm <- CRMetrics$new(cms = testdata.cms, samples = c("sample1", "sample2"), n.cores = 1)

# Create embedding
crm$doPreprocessing()
crm$createEmbedding()

# Plot and extract result
crm$plotFilteredCells(type = "embedding")
filtered.cells <- crm$plotFilteredCells(type = "export")
} else {
message("Package 'conos' not available.")
}
} else {
message("Package 'pagoda2' not available.")
}
}
```

Method `getDepth()`: Extract sequencing depth from Conos object.

Usage:

```
CRMetrics$getDepth(cms = self$cms)
```

Arguments:

`cms` list List of (sparse) count matrices (default = `self$cms`)

Returns: data frame

Examples:

```
\donttest{
if (requireNamespace("pagoda2", quietly = TRUE)) {
if (requireNamespace("conos", quietly = TRUE)) {
# Simulate data
testdata.cms <- lapply(seq_len(2), \x) {
out <- Matrix::rsparsematrix(2e3, 1e3, 0.1)
out[out < 0] <- 1
```

```

dimnames(out) <- list(sapply(seq_len(2e3), \(x) paste0("gene",x)),
sapply(seq_len(1e3), \(x) paste0("cell",x)))
return(out)
})

# Initialize
crm <- CRMetrics$new(cms = testdata.cms, samples = c("sample1", "sample2"), n.cores = 1)

# Create embedding
crm$doPreprocessing()
crm$createEmbedding()

# Get depth
crm$getDepth()
} else {
message("Package 'conos' not available.")
}
} else {
message("Package 'pagoda2' not available.")
}
}
}

```

Method `getMitoFraction()`: Calculate the fraction of mitochondrial genes.

Usage:

```
CRMetrics$getMitoFraction(species = c("human", "mouse"), cms = self$cms)
```

Arguments:

`species` character Species to calculate the mitochondrial fraction for (default = "human").

`cms` list List of (sparse) count matrices (default = `self$cms`)

Returns: data frame

Examples:

```

\donttest{
if (requireNamespace("pagoda2", quietly = TRUE)) {
if (requireNamespace("conos", quietly = TRUE)) {
# Simulate data
testdata.cms <- lapply(seq_len(2), \(x) {
out <- Matrix::rsparsematrix(2e3, 1e3, 0.1)
out[out < 0] <- 1
dimnames(out) <- list(sapply(seq_len(2e3), \(x) paste0("gene",x)),
sapply(seq_len(1e3), \(x) paste0("cell",x)))
return(out)
})

# Initialize
crm <- CRMetrics$new(cms = testdata.cms, samples = c("sample1", "sample2"), n.cores = 1)

# Create embedding
crm$doPreprocessing()

```

```

crm$createEmbedding()

# Get mito. fraction
crm$getMitoFraction(species = c("human", "mouse"))
} else {
message("Package 'conos' not available.")
}
} else {
message("Package 'pagoda2' not available.")
}
}
}

```

Method `prepareCellbender()`: Create plots and script call for CellBender

Usage:

```

CRMetrics$prepareCellbender(
  shrinkage = 100,
  show.expected.cells = TRUE,
  show.total.droplets = TRUE,
  expected.cells = NULL,
  total.droplets = NULL,
  cms.raw = self$cms.raw,
  umi.counts = self$cellbender$umi.counts,
  data.path = self$data.path,
  samples = self$metadata$sample,
  verbose = self$verbose,
  n.cores = self$n.cores,
  unique.names = FALSE,
  sep = "!!"
)

```

Arguments:

`shrinkage` integer Select every nth UMI count per cell for plotting. Improves plotting speed drastically. To plot all cells, set to 1 (default = 100)

`show.expected.cells` logical Plot line depicting expected number of cells (default = TRUE)

`show.total.droplets` logical Plot line depicting total droplets included for CellBender run (default = TRUE)

`expected.cells` named numeric If NULL, expected cells will be deduced from the number of cells per sample identified by Cell Ranger. Otherwise, a named vector of expected cells with sample IDs as names. Sample IDs must match those in `summary.metrics` (default: stored named vector)

`total.droplets` named numeric If NULL, total droplets included will be deduced from expected cells multiplied by 3. Otherwise, a named vector of total droplets included with sample IDs as names. Sample IDs must match those in `summary.metrics` (default: stored named vector)

`cms.raw` list Raw count matrices from HDF5 Cell Ranger outputs (default = `self$cms.raw`)

`umi.counts` list UMI counts calculated as column sums of raw count matrices from HDF5 Cell Ranger outputs (default: stored list)

`data.path` character Path to Cell Ranger outputs (default = `self$data.path`)

samples character Sample names to include (default = self\$metadata\$sample)
 verbose logical Show progress (default: stored vector)
 n.cores integer Number of cores (default: stored vector)
 unique.names logical Create unique cell names (default = FALSE)
 sep character Separator for creating unique cell names (default = "!!")

Returns: ggplot2 object and bash script

Examples:

```

\dontrun{
crm <- CRMetrics$new(data.path = "/path/to/count/data")
crm$prepareCellbender()
}

```

Method saveCellbenderScript():

Usage:

```

CRMetrics$saveCellbenderScript(
  file = "cellbender_script.sh",
  fpr = 0.01,
  epochs = 150,
  use.gpu = TRUE,
  expected.cells = NULL,
  total.droplets = NULL,
  data.path = self$data.path,
  samples = self$metadata$sample,
  args = NULL
)

```

Arguments:

file character File name for CellBender script. Will be stored in data.path (default: "cellbender_script.sh")
 fpr numeric False positive rate for CellBender (default = 0.01)
 epochs integer Number of epochs for CellBender (default = 150)
 use.gpu logical Use CUDA capable GPU (default = TRUE)
 expected.cells named numeric If NULL, expected cells will be deduced from the number of cells per sample identified by Cell Ranger. Otherwise, a named vector of expected cells with sample IDs as names. Sample IDs must match those in summary.metrics (default: stored named vector)
 total.droplets named numeric If NULL, total droplets included will be deduced from expected cells multiplied by 3. Otherwise, a named vector of total droplets included with sample IDs as names. Sample IDs must match those in summary.metrics (default: stored named vector)
 data.path character Path to Cell Ranger outputs (default = self\$data.path)
 samples character Sample names to include (default = self\$metadata\$sample)
 args character (optional) Additional parameters for CellBender

Returns: bash script

Examples:

```

\dontrun{
crm <- CRMetrics$new(data.path = "/path/to/count/data/")
crm$prepareCellbender()
crm$saveCellbenderScript()
}

```

Method `getExpectedCells()`: Extract the expected number of cells per sample based on the Cell Ranger summary metrics

Usage:

```
CRMetrics$getExpectedCells(samples = self$metadata$sample)
```

Arguments:

`samples` character Sample names to include (default = `self$metadata$sample`)

Returns: A numeric vector

Examples:

```

# Simulate data
testdata.cms <- lapply(seq_len(2), \(x) {
  out <- Matrix::rsparsematrix(2e3, 1e3, 0.1)
  out[out < 0] <- 1
  dimnames(out) <- list(sapply(seq_len(2e3), \(x) paste0("gene",x)),
    sapply(seq_len(1e3), \(x) paste0("cell",x)))
  return(out)
})

# Initialize
crm <- CRMetrics$new(cms = testdata.cms, samples = c("sample1", "sample2"), n.cores = 1)

# Get summary
crm$addSummaryFromCms()

# Get no. cells
crm$getExpectedCells()

```

Method `getTotalDroplets()`: Get the total number of droplets included in the CellBender estimations. Based on the Cell Ranger summary metrics and multiplied by a preset multiplier.

Usage:

```
CRMetrics$getTotalDroplets(samples = self$metadata$sample, multiplier = 3)
```

Arguments:

`samples` character Samples names to include (default = `self$metadata$sample`)

`multiplier` numeric Number to multiply expected number of cells with (default = 3)

Returns: A numeric vector

Examples:

```

# Simulate data
testdata.cms <- lapply(seq_len(2), \(x) {
  out <- Matrix::rsparsematrix(2e3, 1e3, 0.1)
  out[out < 0] <- 1

```

```

dimnames(out) <- list(sapply(seq_len(2e3), \(x) paste0("gene",x)),
sapply(seq_len(1e3), \(x) paste0("cell",x)))
return(out)
})

# Initialize
crm <- CRMetrics$new(cms = testdata.cms, samples = c("sample1", "sample2"), n.cores = 1)

# Add summary
crm$addSummaryFromCms()

# Get no. droplets
crm$getTotalDroplets()

```

Method `addCms()`: Add a list of count matrices to the `CRMetrics` object.

Usage:

```

CRMetrics$addCms(
  cms = NULL,
  data.path = self$data.path,
  samples = self$metadata$sample,
  cellbender = FALSE,
  raw = FALSE,
  symbol = TRUE,
  unique.names = TRUE,
  sep = "!!",
  add.metadata = TRUE,
  n.cores = self$n.cores,
  verbose = self$verbose
)

```

Arguments:

`cms` list List of (sparse) count matrices (default = `NULL`)

`data.path` character Path to cellranger count data (default = `self$data.path`).

`samples` character Vector of sample names. If `NULL`, samples are extracted from `cms` (default = `self$metadata$sample`)

`cellbender` logical Add CellBender filtered count matrices in HDF5 format. Requires that "cellbender" is in the names of the files (default = `FALSE`)

`raw` logical Add raw count matrices from Cell Ranger output. Cannot be combined with `cellbender=TRUE` (default = `FALSE`)

`symbol` character The type of gene IDs to use, `SYMBOL` (`TRUE`) or `ENSEMBLE` (default = `TRUE`)

`unique.names` logical Make cell names unique based on `sep` parameter (default = `TRUE`)

`sep` character Separator used to create unique cell names (default = `"!!"`)

`add.metadata` boolean Add metadata from `cms` or not (default = `TRUE`)

`n.cores` integer Number of cores to use (default = `self$n.cores`)

`verbose` boolean Print progress (default = `self$verbose`)

Returns: Add list of (sparse) count matrices to R6 class object

Examples:

```
\dontrun{
crm <- CRMetrics$new(data.path = "/path/to/count/data/")

# Simulate data
testdata.cms <- lapply(seq_len(2), \(x) {
out <- Matrix::rsparsematrix(2e3, 1e3, 0.1)
out[out < 0] <- 1
dimnames(out) <- list(sapply(seq_len(2e3), \(x) paste0("gene",x)),
sapply(seq_len(1e3), \(x) paste0("cell",x)))
return(out)
})

crm$addCms(cms = testdata.cms)
}
```

Method `plotCbTraining()`: Plot the results from the CellBender estimations

Usage:

```
CRMetrics$plotCbTraining(
  data.path = self$data.path,
  samples = self$metadata$sample,
  pal = self$pal
)
```

Arguments:

`data.path` character Path to Cell Ranger outputs (default = `self$data.path`)
`samples` character Sample names to include (default = `self$metadata$sample`)
`pal` character Plotting palette (default = `self$pal`)

Returns: A `ggplot2` object

Examples:

```
\dontrun{
crm <- CRMetrics$new(data.path = "/path/to/count/data/")
crm$prepareCellbender()
crm$saveCellbenderScript()
## Run CellBender script
crm$plotCbTraining()
}
```

Method `plotCbCellProbs()`: Plot the CellBender assigned cell probabilities

Usage:

```
CRMetrics$plotCbCellProbs(
  data.path = self$data.path,
  samples = self$metadata$sample,
  low.col = "gray",
  high.col = "red"
)
```

Arguments:

data.path character Path to Cell Ranger outputs (default = self\$data.path)
 samples character Sample names to include (default = self\$metadata\$sample)
 low.col character Color for low probabilities (default = "gray")
 high.col character Color for high probabilities (default = "red")

Returns: A ggplot2 object

Examples:

```

\dontrun{
crm <- CRMetrics$new(data.path = "/path/to/count/data/")
crm$prepareCellbender()
crm$saveCellbenderScript()
## Run the CellBender script
crm$plotCbCellProbs()
}

```

Method plotCbAmbExp(): Plot the estimated ambient gene expression per sample from CellBender calculations

Usage:

```

CRMetrics$plotCbAmbExp(
  cutoff = 0.005,
  data.path = self$data.path,
  samples = self$metadata$sample
)

```

Arguments:

cutoff numeric Horizontal line included in the plot to indicate highly expressed ambient genes (default = 0.005)
 data.path character Path to Cell Ranger outputs (default = self\$data.path)
 samples character Sample names to include (default = self\$metadata\$sample)

Returns: A ggplot2 object

Examples:

```

\dontrun{
crm <- CRMetrics$new(data.path = "/path/to/count/data/")
crm$prepareCellbender()
crm$saveCellbenderScript()
## Run CellBender script
crm$plotCbAmbExp()
}

```

Method plotCbAmbGenes(): Plot the most abundant estimated ambient genes from the CellBender calculations

Usage:

```

CRMetrics$plotCbAmbGenes(
  cutoff = 0.005,
  data.path = self$data.path,
  samples = self$metadata$sample,
  pal = self$pal
)

```

Arguments:

`cutoff` numeric Cutoff of ambient gene expression to use to extract ambient genes per sample
`data.path` character Path to Cell Ranger outputs (default = `self$data.path`)
`samples` character Sample names to include (default = `self$metadata$sample`)
`pal` character Plotting palette (default = `self$pal`)

Returns: A `ggplot2` object

Examples:

```
\dontrun{
crm <- CRMetrics$new(data.path = "/path/to/count/data/")
crm$prepareCellbender()
crm$saveCellbenderScript()
## Run CellBender script
crm$plotCbAmbGenes()
}
```

Method `addSummaryFromCms()`: Add summary metrics from a list of count matrices

Usage:

```
CRMetrics$addSummaryFromCms(
  cms = self$cms,
  n.cores = self$n.cores,
  verbose = self$verbose
)
```

Arguments:

`cms` list A list of filtered count matrices (default = `self$cms`)
`n.cores` integer Number of cores to use (default = `self$n.cores`)
`verbose` logical Show progress (default = `self$verbose`)

Returns: `data.frame`

Examples:

```
# Simulate data
testdata.cms <- lapply(seq_len(2), \x) {
  out <- Matrix::rsparsematrix(2e3, 1e3, 0.1)
  out[out < 0] <- 1
  dimnames(out) <- list(sapply(seq_len(2e3), \x) paste0("gene",x)),
    sapply(seq_len(1e3), \x) paste0("cell",x))
  return(out)
})

# Initialize
crm <- CRMetrics$new(cms = testdata.cms, samples = c("sample1", "sample2"), n.cores = 1)

# Add summary
crm$addSummaryFromCms()
```

Method `runSoupX()`: Run SoupX ambient RNA estimation and correction

Usage:

```

CRMetrics$runSoupX(
  data.path = self$data.path,
  samples = self$metadata$sample,
  n.cores = self$n.cores,
  verbose = self$verbose,
  arg.load10X = list(),
  arg.autoEstCont = list(),
  arg.adjustCounts = list()
)

```

Arguments:

`data.path` character Path to Cell Ranger outputs (default = `self$data.path`)

`samples` character Sample names to include (default = `self$metadata$sample`)

`n.cores` numeric Number of cores (default = `self$n.cores`)

`verbose` logical Show progress (default = `self$verbose`)

`arg.load10X` list A list with additional parameters for `SoupX::load10X` (default = `list()`)

`arg.autoEstCont` list A list with additional parameters for `SoupX::autoEstCont` (default = `list()`)

`arg.adjustCounts` list A list with additional parameters for `SoupX::adjustCounts` (default = `list()`)

Returns: List containing a list with corrected counts, and a data.frame containing plotting estimations

Examples:

```

\dontrun{
crm <- CRMetrics$new(data.path = "/path/to/count/data/")
crm$runSoupX()
}

```

Method `plotSoupX()`: Plot the results from the `SoupX` estimations

Usage:

```
CRMetrics$plotSoupX(plot.df = self$soupx$plot.df)
```

Arguments:

`plot.df` data.frame `SoupX` estimations (default = `self$soupx$plot.df`)

Returns: A `ggplot2` object

Examples:

```

\dontrun{
crm <- CRMetrics$new(data.path = "/path/to/count/data/")
crm$runSoupX()
crm$plotSoupX()
}

```

Method `plotCbCells()`: Plot CellBender cell estimations against the estimated cell numbers from Cell Ranger

Usage:

```
CRMetrics$plotCbCells(
  data.path = self$data.path,
  samples = self$metadata$sample,
  pal = self$pal
)
```

Arguments:

`data.path` character Path to Cell Ranger outputs (default = `self$data.path`)
`samples` character Sample names to include (default = `self$metadata$sample`)
`pal` character Plotting palette (default = `self$pal`)

Returns: A ggplot2 object

Examples:

```
\dontrun{
crm <- CRMetrics$new(data.path = "/path/to/count/data/")
crm$prepareCellbender()
crm$saveCellbenderScript()
## Run CellBender script
crm$plotCbCells()
}
```

Method `addDoublets()`: Add doublet results created from exported Python script

Usage:

```
CRMetrics$addDoublets(
  method = c("scrublet", "doubletdetection"),
  data.path = self$data.path,
  samples = self$metadata$sample,
  cms = self$cms,
  verbose = self$verbose
)
```

Arguments:

`method` character Which method to use, either `scrublet` or `doubletdetection` (default is both).
`data.path` character Path to Cell Ranger outputs (default = `self$data.path`)
`samples` character Sample names to include (default = `self$metadata$sample`)
`cms` list List containing the count matrices (default = `self$cms`).
`verbose` boolean Print progress (default = `self$verbose`)

Returns: List of doublet results

Examples:

```
\dontrun{
crm <- CRMetrics$new(data.path = "/path/to/count/data/")
crm$detectDoublets(export = TRUE)
## Run Python script
crm$addDoublets()
}
```

Method `clone()`: The objects of this class are cloneable with this method.

Usage:

```
CRMetrics$clone(deep = FALSE)
```

Arguments:

```
deep Whether to make a deep clone.
```

Examples

```
## -----
## Method `CRMetrics$new`
## -----

## Not run:
crm <- CRMetrics$new(data.path = "/path/to/count/data/")

## End(Not run)

## -----
## Method `CRMetrics$addDetailedMetrics`
## -----

# Simulate data
testdata.cms <- lapply(seq_len(2), \(x) {
  out <- Matrix::rsparsematrix(2e3, 1e3, 0.1)
  out[out < 0] <- 1
  dimnames(out) <- list(sapply(seq_len(2e3), \(x) paste0("gene",x)),
    sapply(seq_len(1e3), \(x) paste0("cell",x)))
  return(out)
})

# Initialize
crm <- CRMetrics$new(cms = testdata.cms, samples = c("sample1", "sample2"), n.cores = 1)

# Run function
crm$addDetailedMetrics()

## -----
## Method `CRMetrics$addComparison`
## -----

# Simulate data
testdata.cms <- lapply(seq_len(2), \(x) {
  out <- Matrix::rsparsematrix(2e3, 1e3, 0.1)
  out[out < 0] <- 1
  dimnames(out) <- list(sapply(seq_len(2e3), \(x) paste0("gene",x)),
    sapply(seq_len(1e3), \(x) paste0("cell",x)))
  return(out)
})

# Initialize
crm <- CRMetrics$new(cms = testdata.cms, samples = c("sample1", "sample2"), n.cores = 1)

# Add metadata
```

```

crm$metadata <- data.frame(sex = c("male","female"))

# Add comparison group
crm$addComparison(comp.group = "sex")

## -----
## Method `CRMetric$plotSamples`
## -----

samples <- c("sample1", "sample2")

# Simulate data
testdata.cms <- lapply(seq_len(2), \(\x) {
  out <- Matrix::rsparsematrix(2e3, 1e3, 0.1)
  out[out < 0] <- 1
  dimnames(out) <- list(sapply(seq_len(2e3), \(\x) paste0("gene",x)),
    sapply(seq_len(1e3), \(\x) paste0("cell",x)))
  return(out)
})
names(testdata.cms) <- samples

# Create metadata
metadata <- data.frame(sample = samples,
  sex = c("male","female"),
  condition = c("a","b"))

# Initialize
crm <- CRMetric$new(cms = testdata.cms, metadata = metadata, n.cores = 1)

# Plot
crm$plotSamples(comp.group = "sex", second.comp.group = "condition")

## -----
## Method `CRMetric$plotSummaryMetrics`
## -----

# Simulate data
testdata.cms <- lapply(seq_len(2), \(\x) {
  out <- Matrix::rsparsematrix(2e3, 1e3, 0.1)
  out[out < 0] <- 1
  dimnames(out) <- list(sapply(seq_len(2e3), \(\x) paste0("gene",x)),
    sapply(seq_len(1e3), \(\x) paste0("cell",x)))
  return(out)
})

# Initialize
crm <- CRMetric$new(cms = testdata.cms, samples = c("sample1", "sample2"), n.cores = 1)

# Add summary metrics
crm$addSummaryFromCms()

crm$plotSummaryMetrics(plot.geom = "point")

```

```

## -----
## Method `CRMetrics$plotDetailedMetrics`
## -----

# Simulate data
testdata.cms <- lapply(seq_len(2), \(x) {
  out <- Matrix::rsparsematrix(2e3, 1e3, 0.1)
  out[out < 0] <- 1
  dimnames(out) <- list(sapply(seq_len(2e3), \(x) paste0("gene",x)),
    sapply(seq_len(1e3), \(x) paste0("cell",x)))
  return(out)
})

# Initialize
crm <- CRMetrics$new(cms = testdata.cms, samples = c("sample1", "sample2"), n.cores = 1)

# Add detailed metrics
crm$addDetailedMetrics()

# Plot
crm$plotDetailedMetrics()

## -----
## Method `CRMetrics$plotEmbedding`
## -----

if (requireNamespace("pagoda2", quietly = TRUE)) {
  if (requireNamespace("conos", quietly = TRUE)) {
    # Simulate data
    testdata.cms <- lapply(seq_len(2), \(x) {
      out <- Matrix::rsparsematrix(2e3, 1e3, 0.1)
      out[out < 0] <- 1
      dimnames(out) <- list(sapply(seq_len(2e3), \(x) paste0("gene",x)),
        sapply(seq_len(1e3), \(x) paste0("cell",x)))
      return(out)
    })

    # Initialize
    crm <- CRMetrics$new(cms = testdata.cms, samples = c("sample1", "sample2"), n.cores = 1)

    # Create embedding
    crm$doPreprocessing()
    crm$createEmbedding()

    crm$plotEmbedding()
  } else {
    message("Package 'conos' not available.")
  }
}

```

```

} else {
message("Package 'pagoda2' not available.")
}

## -----
## Method `CRMetric$plotDepth`
## -----

if (requireNamespace("pagoda2", quietly = TRUE)) {
if (requireNamespace("conos", quietly = TRUE)) {
# Simulate data
testdata.cms <- lapply(seq_len(2), \x) {
out <- Matrix::rsparsematrix(2e3, 1e3, 0.1)
out[out < 0] <- 1
dimnames(out) <- list(sapply(seq_len(2e3), \x) paste0("gene",x)),
sapply(seq_len(1e3), \x) paste0("cell",x))
return(out)
})

# Initialize
crm <- CRMetric$new(cms = testdata.cms, samples = c("sample1", "sample2"), n.cores = 1)

# Create embedding
crm$doPreprocessing()
crm$createEmbedding()

# Plot
crm$plotDepth()
} else {
message("Package 'conos' not available.")
}
} else {
message("Package 'pagoda2' not available.")
}

## -----
## Method `CRMetric$plotMitoFraction`
## -----

if (requireNamespace("pagoda2", quietly = TRUE)) {
if (requireNamespace("conos", quietly = TRUE)) {
# Simulate data
testdata.cms <- lapply(seq_len(2), \x) {
out <- Matrix::rsparsematrix(2e3, 1e3, 0.1)
out[out < 0] <- 1
dimnames(out) <- list(sapply(seq_len(2e3), \x) paste0("gene",x)),
sapply(seq_len(1e3), \x) paste0("cell",x))
return(out)
})
}
}

```



```

# Initialize
crm <- CRMetrics$new(cms = testdata.cms, samples = c("sample1", "sample2"), n.cores = 1)

# Create embedding
crm$doPreprocessing()
crm$createEmbedding()

# Plot
crm$plotMitoFraction()
} else {
message("Package 'conos' not available.")
}
} else {
message("Package 'pagoda2' not available.")
}

## -----
## Method `CRMetrics$detectDoublets`
## -----

## Not run:
# Simulate data
testdata.cms <- lapply(seq_len(2), \(x) {
out <- Matrix::rsparsematrix(2e3, 1e3, 0.1)
out[out < 0] <- 1
dimnames(out) <- list(sapply(seq_len(2e3), \(x) paste0("gene",x)),
sapply(seq_len(1e3), \(x) paste0("cell",x)))
return(out)
})

# Initialize
crm <- CRMetrics$new(cms = testdata.cms, samples = c("sample1", "sample2"), n.cores = 1)

# Detect doublets
crm$detectDoublets(method = "scrublet",
conda.path = "/opt/software/miniconda/4.12.0/condabin/conda")

## End(Not run)

## -----
## Method `CRMetrics$doPreprocessing`
## -----

if (requireNamespace("pagoda2", quietly = TRUE)) {
# Simulate data
testdata.cms <- lapply(seq_len(2), \(x) {
out <- Matrix::rsparsematrix(2e3, 1e3, 0.1)
out[out < 0] <- 1
dimnames(out) <- list(sapply(seq_len(2e3), \(x) paste0("gene",x)),

```

```

sapply(seq_len(1e3), \ (x) paste0("cell",x)))
return(out)
})

# Initialize
crm <- CRMetric$new(cms = testdata.cms, samples = c("sample1", "sample2"), n.cores = 1)

# Perform preprocessing
crm$doPreprocessing(preprocess = "pagoda2")
} else {
message("Package 'pagoda2' not available.")
}

## -----
## Method `CRMetric$createEmbedding`
## -----

if (requireNamespace("pagoda2", quietly = TRUE)) {
if (requireNamespace("conos", quietly = TRUE)) {
# Simulate data
testdata.cms <- lapply(seq_len(2), \ (x) {
out <- Matrix::rsparsematrix(2e3, 1e3, 0.1)
out[out < 0] <- 1
dimnames(out) <- list(sapply(seq_len(2e3), \ (x) paste0("gene",x)),
sapply(seq_len(1e3), \ (x) paste0("cell",x)))
return(out)
})

# Initialize
crm <- CRMetric$new(cms = testdata.cms, samples = c("sample1", "sample2"), n.cores = 1)

# Create embedding
crm$doPreprocessing()
crm$createEmbedding()
} else {
message("Package 'conos' not available.")
}
} else {
message("Package 'pagoda2' not available.")
}

## -----
## Method `CRMetric$filterCms`
## -----

if (requireNamespace("pagoda2", quietly = TRUE)) {
if (requireNamespace("conos", quietly = TRUE)) {
# Simulate data
testdata.cms <- lapply(seq_len(2), \ (x) {

```

```

out <- Matrix::rsparsematrix(2e3, 1e3, 0.1)
out[out < 0] <- 1
dimnames(out) <- list(sapply(seq_len(2e3), \(x) paste0("gene",x)),
  sapply(seq_len(1e3), \(x) paste0("cell",x)))
return(out)
})

# Initialize
crm <- CRMetrics$new(cms = testdata.cms, samples = c("sample1", "sample2"), n.cores = 1)

# Create embedding
crm$doPreprocessing()
crm$createEmbedding()

# Filter CMs
crm$filterCms(depth.cutoff = 1e3, mito.cutoff = 0.05)
} else {
message("Package 'conos' not available.")
}
} else {
message("Package 'pagoda2' not available.")
}

## -----
## Method `CRMetrics$selectMetrics`
## -----

# Simulate data
testdata.cms <- lapply(seq_len(2), \(x) {
out <- Matrix::rsparsematrix(2e3, 1e3, 0.1)
out[out < 0] <- 1
dimnames(out) <- list(sapply(seq_len(2e3), \(x) paste0("gene",x)),
  sapply(seq_len(1e3), \(x) paste0("cell",x)))
return(out)
})

# Initialize
crm <- CRMetrics$new(cms = testdata.cms, samples = c("sample1", "sample2"), n.cores = 1)

# Select metrics
crm$selectMetrics()
selection.metrics <- crm$selectMetrics(c(1:4))

## -----
## Method `CRMetrics$plotFilteredCells`
## -----

if (requireNamespace("pagoda2", quietly = TRUE)) {
if (requireNamespace("conos", quietly = TRUE)) {
# Simulate data

```

```

testdata.cms <- lapply(seq_len(2), \(x) {
  out <- Matrix::rsparsematrix(2e3, 1e3, 0.1)
  out[out < 0] <- 1
  dimnames(out) <- list(sapply(seq_len(2e3), \(x) paste0("gene",x)),
    sapply(seq_len(1e3), \(x) paste0("cell",x)))
  return(out)
})

# Initialize
crm <- CRMetrics$new(cms = testdata.cms, samples = c("sample1", "sample2"), n.cores = 1)

# Create embedding
crm$doPreprocessing()
crm$createEmbedding()

# Plot and extract result
crm$plotFilteredCells(type = "embedding")
filtered.cells <- crm$plotFilteredCells(type = "export")
} else {
  message("Package 'conos' not available.")
}
} else {
  message("Package 'pagoda2' not available.")
}

## -----
## Method `CRMetrics$getDepth`
## -----

if (requireNamespace("pagoda2", quietly = TRUE)) {
  if (requireNamespace("conos", quietly = TRUE)) {
    # Simulate data
    testdata.cms <- lapply(seq_len(2), \(x) {
      out <- Matrix::rsparsematrix(2e3, 1e3, 0.1)
      out[out < 0] <- 1
      dimnames(out) <- list(sapply(seq_len(2e3), \(x) paste0("gene",x)),
        sapply(seq_len(1e3), \(x) paste0("cell",x)))
      return(out)
    })

    # Initialize
    crm <- CRMetrics$new(cms = testdata.cms, samples = c("sample1", "sample2"), n.cores = 1)

    # Create embedding
    crm$doPreprocessing()
    crm$createEmbedding()

    # Get depth
    crm$getDepth()
  } else {
    message("Package 'conos' not available.")
  }
}

```

```

}
} else {
message("Package 'pagoda2' not available.")
}

## -----
## Method `CRMetrics$getMitoFraction`
## -----

if (requireNamespace("pagoda2", quietly = TRUE)) {
if (requireNamespace("conos", quietly = TRUE)) {
# Simulate data
testdata.cms <- lapply(seq_len(2), \x) {
out <- Matrix::rsparsematrix(2e3, 1e3, 0.1)
out[out < 0] <- 1
dimnames(out) <- list(sapply(seq_len(2e3), \x) paste0("gene",x)),
sapply(seq_len(1e3), \x) paste0("cell",x))
return(out)
})

# Initialize
crm <- CRMetrics$new(cms = testdata.cms, samples = c("sample1", "sample2"), n.cores = 1)

# Create embedding
crm$doPreprocessing()
crm$createEmbedding()

# Get mito. fraction
crm$getMitoFraction(species = c("human", "mouse"))
} else {
message("Package 'conos' not available.")
}
} else {
message("Package 'pagoda2' not available.")
}

## -----
## Method `CRMetrics$prepareCellbender`
## -----

## Not run:
crm <- CRMetrics$new(data.path = "/path/to/count/data")
crm$prepareCellbender()

## End(Not run)

## -----
## Method `CRMetrics$saveCellbenderScript`
## -----

```

```

## Not run:
crm <- CRMetrics$new(data.path = "/path/to/count/data/")
crm$prepareCellbender()
crm$saveCellbenderScript()

## End(Not run)

## -----
## Method `CRMetrics$getExpectedCells`
## -----

# Simulate data
testdata.cms <- lapply(seq_len(2), \(x) {
  out <- Matrix::rsparsematrix(2e3, 1e3, 0.1)
  out[out < 0] <- 1
  dimnames(out) <- list(sapply(seq_len(2e3), \(x) paste0("gene",x)),
    sapply(seq_len(1e3), \(x) paste0("cell",x)))
  return(out)
})

# Initialize
crm <- CRMetrics$new(cms = testdata.cms, samples = c("sample1", "sample2"), n.cores = 1)

# Get summary
crm$addSummaryFromCms()

# Get no. cells
crm$getExpectedCells()

## -----
## Method `CRMetrics$getTotalDroplets`
## -----

# Simulate data
testdata.cms <- lapply(seq_len(2), \(x) {
  out <- Matrix::rsparsematrix(2e3, 1e3, 0.1)
  out[out < 0] <- 1
  dimnames(out) <- list(sapply(seq_len(2e3), \(x) paste0("gene",x)),
    sapply(seq_len(1e3), \(x) paste0("cell",x)))
  return(out)
})

# Initialize
crm <- CRMetrics$new(cms = testdata.cms, samples = c("sample1", "sample2"), n.cores = 1)

# Add summary
crm$addSummaryFromCms()

# Get no. droplets
crm$getTotalDroplets()

## -----
## Method `CRMetrics$addCms`

```

```

## -----

## Not run:
crm <- CRMetrics$new(data.path = "/path/to/count/data/")

# Simulate data
testdata.cms <- lapply(seq_len(2), \(x) {
  out <- Matrix::rsparsematrix(2e3, 1e3, 0.1)
  out[out < 0] <- 1
  dimnames(out) <- list(sapply(seq_len(2e3), \(x) paste0("gene",x)),
    sapply(seq_len(1e3), \(x) paste0("cell",x)))
  return(out)
})

crm$addCms(cms = testdata.cms)

## End(Not run)

## -----
## Method `CRMetrics$plotCbTraining`
## -----

## Not run:
crm <- CRMetrics$new(data.path = "/path/to/count/data/")
crm$prepareCellbender()
crm$saveCellbenderScript()
## Run CellBender script
crm$plotCbTraining()

## End(Not run)

## -----
## Method `CRMetrics$plotCbCellProbs`
## -----

## Not run:
crm <- CRMetrics$new(data.path = "/path/to/count/data/")
crm$prepareCellbender()
crm$saveCellbenderScript()
## Run the CellBender script
crm$plotCbCellProbs()

## End(Not run)

## -----
## Method `CRMetrics$plotCbAmbExp`
## -----

## Not run:
crm <- CRMetrics$new(data.path = "/path/to/count/data/")
crm$prepareCellbender()
crm$saveCellbenderScript()
## Run CellBender script

```

```

crm$plotCbAmbExp()

## End(Not run)

## -----
## Method `CRMetrics$plotCbAmbGenes`
## -----

## Not run:
crm <- CRMetrics$new(data.path = "/path/to/count/data/")
crm$prepareCellbender()
crm$saveCellbenderScript()
## Run CellBender script
crm$plotCbAmbGenes()

## End(Not run)

## -----
## Method `CRMetrics$addSummaryFromCms`
## -----

# Simulate data
testdata.cms <- lapply(seq_len(2), \(x) {
  out <- Matrix::rsparsematrix(2e3, 1e3, 0.1)
  out[out < 0] <- 1
  dimnames(out) <- list(sapply(seq_len(2e3), \(x) paste0("gene",x)),
    sapply(seq_len(1e3), \(x) paste0("cell",x)))
  return(out)
})

# Initialize
crm <- CRMetrics$new(cms = testdata.cms, samples = c("sample1", "sample2"), n.cores = 1)

# Add summary
crm$addSummaryFromCms()

## -----
## Method `CRMetrics$runSoupX`
## -----

## Not run:
crm <- CRMetrics$new(data.path = "/path/to/count/data/")
crm$runSoupX()

## End(Not run)

## -----
## Method `CRMetrics$plotSoupX`
## -----

## Not run:
crm <- CRMetrics$new(data.path = "/path/to/count/data/")
crm$runSoupX()

```



```

crm$plotSoupX()

## End(Not run)

## -----
## Method `CRMetrics$plotCbCells`
## -----

## Not run:
crm <- CRMetrics$new(data.path = "/path/to/count/data/")
crm$prepareCellbender()
crm$saveCellbenderScript()
## Run CellBender script
crm$plotCbCells()

## End(Not run)

## -----
## Method `CRMetrics$addDoublets`
## -----

## Not run:
crm <- CRMetrics$new(data.path = "/path/to/count/data/")
crm$detectDoublets(export = TRUE)
## Run Python script
crm$addDoublets()

## End(Not run)

```

read10xH5

Read 10x HDF5 files

Description

Read 10x HDF5 files

Usage

```

read10xH5(
  data.path,
  samples = NULL,
  type = c("raw", "filtered", "cellbender", "cellbender_filtered"),
  symbol = TRUE,
  sep = "!!",
  n.cores = 1,
  verbose = TRUE,
  unique.names = FALSE
)

```

Arguments

<code>data.path</code>	character
<code>samples</code>	character vector, select specific samples for processing (default = NULL)
<code>type</code>	name of H5 file to search for, "raw" and "filtered" are Cell Ranger count outputs, "cellbender" is output from CellBender after running script from saveCellbenderScript
<code>symbol</code>	logical Use gene SYMBOLs (TRUE) or ENSEMBL IDs (FALSE) (default = TRUE)
<code>sep</code>	character Separator for creating unique cell names from sample IDs and cell IDs (default = "!!")
<code>n.cores</code>	integer Number of cores (default = 1)
<code>verbose</code>	logical Print progress (default = TRUE)
<code>unique.names</code>	logical Create unique cell IDs (default = FALSE)

Value

list with sparse count matrices

Examples

```
## Not run:  
cms.h5 <- read10xH5(data.path = "/path/to/count/data")  
  
## End(Not run)
```

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