## Package 'VisiumIO'

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Title Import Visium data from the 10X Space Ranger pipeline

Version 1.3.2

**Description** The package allows users to readily import spatial data obtained from either the 10X website or from the Space Ranger pipeline. Supported formats include tar.gz, h5, and mtx files. Multiple files can be imported at once with \*List type of functions. The package represents data mainly as SpatialExperiment objects.

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**Depends** R ( $\geq$  4.4.0), TENxIO

- Imports BiocBaseUtils, BiocGenerics, BiocIO (>= 1.15.1), jsonlite, methods, S4Vectors, SingleCellExperiment, SpatialExperiment, SummarizedExperiment
- Suggests arrow, BiocStyle, data.table, knitr, readr, rmarkdown, tinytest
- biocViews Software, Infrastructure, DataImport, SingleCell, Spatial

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BugReports https://github.com/waldronlab/VisiumIO/issues

#### URL https://github.com/waldronlab/VisiumIO

Collate 'TENxSpatialCSV.R' 'TENxSpatialList-class.R' 'TENxSpatialParquet.R' 'TENxVisium-class.R' 'TENxVisiumList-class.R' 'TENxVisiumHD-class.R' 'utilities.R' Date 2024-12-05 git\_url https://git.bioconductor.org/packages/VisiumIO git\_branch devel git\_last\_commit\_3588697 git\_last\_commit\_date 2024-12-05

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compareBarcodes Compare barcodes between raw and filtered data

#### Description

This function compares the barcodes between raw and filtered data **depending** on the order of processing. Typically, the "raw" barcodes are compared to the "filtered" ones. The presence of raw barcodes in the filtered data are marked as TRUE in the resulting data.frame.

## Usage

```
compareBarcodes(
  from_resource,
  to_resource,
  spacerangerOut,
  format = c("mtx", "h5"),
  processing = c("raw", "filtered"),
   ...
)
```

from_resource	character(1) The path to the resource file whose barcodes are used as the basis of the comparison; typically, the "raw" feature barcodes are used.
to_resource	character(1) The path to the resource file whose barcodes are compared to the from_resource; typically, the "filtered" feature barcodes.

spaceranger0ut	character(1) A single string specifying the path to the sample directory of spaceranger count. The directory must contain the (processing)_feature_bc_matrix and spatial sub directories in addition to the outs folder.
format	The format of the output. If missing and con is a file name, the format is derived from the file extension. This argument is unnecessary when con is a derivative of BiocFile.
processing	<pre>character(2) A vector of length 2 that corresponds to the processing type. The processing types are typically "raw" and "filtered". These are the prefixes of the folder names raw_feature_bc_matrix and filtered_feature_bc_matrix. The order of the vector determines the comparison. By default, processing = c("raw", "filtered"), which means barcodes in the raw data are compared to the filtered data.</pre>
	Additional arguments passed to TENxH5 or TENxFileList.

#### Value

A data.frame with barcodes of the first element in the processing data type as the first column and a logical vector indicating whether the barcodes are found in the second element in processing. For example, if processing is c("raw", "filtered"), then the first column will be the barcodes in the raw data and the second column will be a logical vector indicating whether the barcodes are found in the filtered data.

```
if (interactive()) {
    compareBarcodes(
       from_resource = "V1_Adult_Mouse_Brain_raw_feature_bc_matrix.tar.gz",
        to_resource =
            "V1_Adult_Mouse_Brain_filtered_feature_bc_matrix.tar.gz",
   )
    compareBarcodes(
       from_resource =
            "V1_Adult_Mouse_Brain_raw_feature_bc_matrix.h5",
       to_resource =
            "V1_Adult_Mouse_Brain_filtered_feature_bc_matrix.h5"
   )
   compareBarcodes(spacerangerOut = "~/data/outs", format = "h5")
   compareBarcodes(
        spacerangerOut = "~/data/feature_bc_matrix", format = "mtx"
    )
   compareBarcodes(
       spacerangerOut = "~/data/folder_feature_bc_matrix", format = "mtx"
   )
}
```

TENxSpatialCSV-class Represent and import spatial CSV data from 10X Genomics

#### Description

TENxSpatialCSV is a class to represent and import spatial CSV files with specific column names. It is a composed class of TENxIO::TENxFile and contains additional slots for the column names and whether the CSV is a list-type of file.

#### Usage

TENxSpatialCSV(resource, colnames = .TISSUE\_POS\_COLS)

## S4 method for signature 'TENxSpatialCSV,ANY,ANY'
import(con, format, text, ...)

#### Arguments

resource	character(1) The path to the file
colnames	<pre>character() A vector specifying the column names of the CSV, defaults to c("barcode", "in_tissue", "array_row", "array_col", "pxl_row_in_fullres", "pxl_col_in_fullres"). Mainly used for the "positions" CSV type of file which does not include column names in the file.</pre>
con	The connection from which data is loaded or to which data is saved. If this is a character vector, it is assumed to be a file name and a corresponding file connection is created and then closed after exporting the object. If it is a <b>BiocFile</b> derivative, the data is loaded from or saved to the underlying resource. If missing, the function will return the output as a character vector, rather than writing to a connection.
format	The format of the output. If missing and con is a file name, the format is derived from the file extension. This argument is unnecessary when con is a derivative of BiocFile.
text	If con is missing, this can be a character vector directly providing the string data to import.
	Additional inputs to the low level class generator functions

#### Details

Typically, the user will not create an object of this class directly but rather use the TENxVisium() constructor function to create an object of this class in the background. The column names are set to the default values of c("barcode", "in\_tissue", "array\_row", "array\_col", "pxl\_row\_in\_fullres", "pxl\_col\_in\_fullres"). The column names can be changed by specifying the colnames argument in the constructor function.

Set the option "VisiumIO.csvreader" to either "data.table" or "readr" to use the data.table::fread or readr::read\_csv functions, respectively. These options are useful when the CSV file is relatively large and the user wants to use faster read-in options. Note that the outputs will still be converted to DataFrame when incorporated to the SpatialExperiment or SingleCellExperiment object.

#### Value

TENxSpatialCSV: An object of class TENxSpatialCSV

import-method: A DataFrame object containing the data from the CSV file

#### Slots

isList logical(1) A scalar specifying whether the CSV is a list-type of file

colnames character() A vector specifying the column names of the CSV

- variant character(1) A scalar specifying the variant of the CSV file "positions", "cell\_boundaries", or "other". The variant is determined by the name of the CSV file within the constructor function. Values include "positions", "cell\_boundaries", and "other".
- compressed logical(1) A scalar specifying whether the CSV is compressed (mainly with a .gz file extension).

#### Examples

```
sample_dir <- system.file(
    file.path("extdata", "10xVisium", "section1"),
    package = "SpatialExperiment"
)
spatial_dir <- Filter(
    function(x) endsWith(x, "spatial"), list.dirs(sample_dir)
)
csvresource <- file.path(spatial_dir, "tissue_positions_list.csv")
TENxSpatialCSV(csvresource)
head(import(TENxSpatialCSV(csvresource)), 4)
import(TENxSpatialCSV(csvresource)) |>
    attr("metadata") |>
    lapply(names)
```

TENxSpatialList-class A class to represent and import spatial Visium data

#### Description

This class is a composed class of TENxFileList, which can contain a list of TENxFile objects, and a TENxSpatialList object. It is meant to handle spatial Visium data from 10X Genomics.

## Usage

```
TENxSpatialList(
  resources,
  sample_id = "sample01",
  images = c("lowres", "hires", "detected", "aligned", "aligned_fiducials"),
  jsonFile = .SCALE_JSON_FILE,
  tissuePattern = "tissue_positions.*",
  bin_size = character(0L),
  ...
)
```

```
## S4 method for signature 'TENxSpatialList,ANY,ANY'
import(con, format, text, ...)
```

## Arguments

resources	A TENxFileList object or a file path to the tarball containing the matrix / assay data resources.
sample_id	character(1) A single string specifying the sample ID.
images	character() A vector specifying the images to be imported; can be one or multiple of "lowres", "hires", "detected", "aligned".
jsonFile	character(1) A single string specifying the name of the JSON file containing the scale factors.
tissuePattern	character(1) A single string specifying the pattern to match the tissue positions file.
bin_size	character(1) The bin size of the images to import. The default is 008. It corresponds to the directory name square_000um where 000 is the bin value.
	Parameters to pass to the format-specific method.
con	The connection from which data is loaded or to which data is saved. If this is a character vector, it is assumed to be a file name and a corresponding file connection is created and then closed after exporting the object. If it is a <b>BiocFile</b> derivative, the data is loaded from or saved to the underlying resource. If missing, the function will return the output as a character vector, rather than writing to a connection.
format	The format of the output. If missing and con is a file name, the format is derived from the file extension. This argument is unnecessary when con is a derivative of BiocFile.
text	If con is missing, this can be a character vector directly providing the string data to import.

## Details

Typically, the user will not create an object of this class directly but rather use the TENxVisium() constructor function to create an object of this class.

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#### Value

A SpatialExperiment object

#### Methods (by generic)

 import(con = TENxSpatialList, format = ANY, text = ANY): Import a TENxSpatialList object

#### Slots

images character() The image names to use with grep and include in the list of files.

- scaleJSON character(1) The file name of the scale factors JSON file, defaults to 'scalefactors\_json.json'.
- tissuePos character(1) The file name of the tissue positions file; typically a .parquet or .csv file.
- sampleId character(1) A scalar specifying the sample identifier.
- binSize The bin size of the images to import. The default slot value is character(). It typically corresponds to the directory name square\_000um where 000 is the bin value.

## Examples

```
spatial_dir <- system.file(
    file.path("extdata", "10xVisium", "section1", "outs", "spatial"),
    package = "SpatialExperiment"
)
TENxSpatialList(resources = spatial_dir, images = "lowres")
TENxSpatialList(resources = spatial_dir, images = "lowres") |>
    metadata() |> lapply(names)
```

TENxSpatialParquet-class

Represent and import spatial Parquet data from 10X Genomics

#### Description

TENxSpatialParquet is a class to represent and import spatial Parquet files with specific column names. It is a composed class of TENxIO::TENxFile and contains additional slots for the column names and whether the Parquet is a list-type of file.

#### Usage

```
TENxSpatialParquet(resource, colnames = .TISSUE_POS_COLS)
## S4 method for signature 'TENxSpatialParquet,ANY,ANY'
import(con, format, text, ...)
```

#### Arguments

resource	character(1) The path to the file
colnames	<pre>character() A vector specifying the column names of the Parquet, defaults to c("barcode", "in_tissue", "array_row", "array_col", "pxl_row_in_fullres", "pxl_col_in_fullres").</pre>
con	The connection from which data is loaded or to which data is saved. If this is a character vector, it is assumed to be a file name and a corresponding file connection is created and then closed after exporting the object. If it is a BiocFile derivative, the data is loaded from or saved to the underlying resource. If missing, the function will return the output as a character vector, rather than writing to a connection.
format	The format of the output. If missing and con is a file name, the format is derived from the file extension. This argument is unnecessary when con is a derivative of BiocFile.
text	If con is missing, this can be a character vector directly providing the string data to import.
	Additional inputs to the low level class generator functions

#### Details

Typically, the user will not create an object of this class directly but rather use the TENxVisium() constructor function to create an object of this class in the background. The column names are set to the default values of c("barcode", "in\_tissue", "array\_row", "array\_col", "pxl\_row\_in\_fullres", "pxl\_col\_in\_fullres"). The column names can be changed by specifying the colnames argument in the constructor function.

#### Value

TENxSpatialParquet(): An object of class TENxSpatialParquet

import-method: A DataFrame object containing the data from the Parquet file

#### Slots

colnames character() A vector specifying the column names of the Parquet

```
sample_dir <- system.file(
    file.path("extdata", "binned_outputs", "square_002um", "spatial"),
    package = "VisiumI0"
)
spatial_dir <- Filter(
    function(x) endsWith(x, "spatial"), list.dirs(sample_dir)
)
parquetres <- file.path(spatial_dir, "tissue_positions.parquet")
TENxSpatialParquet(parquetres)
import(TENxSpatialParquet(parquetres))</pre>
```

```
## metadata in attributes
import(TENxSpatialParquet(parquetres)) |>
    attr("metadata") |>
    lapply(names)
```

TENxVisium-class A class to represent and import a single Visium Sample

#### Description

This class is a composed class of TENxFileList which can contain a list of TENxFile objects and a TENxSpatialList object. It is meant to handle a single Visium sample from 10X Genomics.

#### Usage

```
TENxVisium(
  resources,
  spatialResource,
  spacerangerOut,
  sample_id = "sample01",
  processing = c("filtered", "raw"),
  format = c("mtx", "h5"),
  images = c("lowres", "hires", "detected", "aligned"),
  jsonFile = .SCALE_JSON_FILE,
  tissuePattern = "tissue_positions.*\\.csv",
  spatialCoordsNames = c("pxl_col_in_fullres", "pxl_row_in_fullres"),
  ...
)
```

## S4 method for signature 'TENxVisium,ANY,ANY'
import(con, format, text, ...)

resources	A TENxFileList object or a file path to the tarball containing the matrix / assay
	data resources.
spatialResource	
	A TENxSpatialList object or a file path to the tarball containing the spatial data.
spacerangerOut	character(1) A single string specifying the path to the sample directory of spaceranger count. The directory must contain the (processing)_feature_bc_matrix and spatial sub directories in addition to the outs folder.
sample_id	character(1) A single string specifying the sample ID.
processing	character(1) A single string indicating the processing folder available e.g., "filtered_feature_barcode_matrix" in the spacerangerOut folder. It can be ei- ther "filtered" or "raw" (default "filtered"). Only used when spacerangerOut is specified.

format	The format of the output. If missing and con is a file name, the format is derived from the file extension. This argument is unnecessary when con is a derivative of BiocFile.
images	character() A vector specifying the images to be imported; can be one or multiple of "lowres", "hires", "detected", "aligned".
jsonFile	character(1) A single string specifying the name of the JSON file containing the scale factors.
tissuePattern	character(1) A single string specifying the pattern to match the tissue positions file.
spatialCoordsNa	mes
	character() A vector of strings specifying the names of the columns in the spatial data containing the spatial coordinates.
	In the constructor, additional arguments passed to TENxFileList(); otherwise, not used.
con	The connection from which data is loaded or to which data is saved. If this is a character vector, it is assumed to be a file name and a corresponding file connection is created and then closed after exporting the object. If it is a BiocFile derivative, the data is loaded from or saved to the underlying resource. If missing, the function will return the output as a character vector, rather than writing to a connection.
text	If con is missing, this can be a character vector directly providing the string data to import.

#### Details

Typically, the user will not create an object of this class directly but rather use TENxVisiumList constructor function for multiple samples. Note that the images, jsonFile, tissuePattern, and spatialCoordsNames arguments are only considered when the spacerangerOut argument or both the resources and spatialResource arguments are paths to files.

#### Value

A SpatialExperiment object

#### Functions

• import(con = TENxVisium, format = ANY, text = ANY): Import Visium data

#### Slots

resources A TENxFileList or TENxH5 object containing the Visium data.

spatialList A TENxSpatialList object containing the spatial

coordNames character() A vector specifying the names of the columns in the spatial data containing the spatial coordinates.

sampleId character(1) A scalar specifying the sample identifier.

#### TENx Visium-class

## See Also

https://support.10xgenomics.com/spatial-gene-expression/software/pipelines/latest/ output/overview

```
sample_dir <- system.file(</pre>
    file.path("extdata", "10xVisium", "section1"),
    package = "VisiumIO"
)
## using spacerangerOut folder
tv <- TENxVisium(</pre>
    spacerangerOut = sample_dir, processing = "raw", images = "lowres"
)
import(tv)
## with TENxFileList spacerangerOut input
tvfl <- TENxVisium(</pre>
    spacerangerOut = TENxFileList(sample_dir),
    format = "mtx",
    processing = "raw",
    images = "lowres"
)
import(tvfl)
## check metadata of the object
import(tvfl) |>
    metadata() |>
    lapply(names)
## importing h5 format
tvfl <- TENxVisium(</pre>
    spacerangerOut = sample_dir,
    format = "h5",
    processing = "raw",
    images = "lowres"
)
import(tvfl)
rffolder <- file.path(sample_dir, "outs", "raw_feature_bc_matrix")</pre>
## using resources and spatialResource inputs
tvfl <- TENxVisium(</pre>
    resources = rffolder,
    spatialResource = file.path(dirname(rffolder), "spatial"),
    format = "mtx",
    processing = "raw",
    images = "lowres"
)
```

```
import(tvfl)
```

TENxVisiumHD-class A class to represent and import multiple Visium HD samples

#### Description

This class contains a SimpleList of TENxVisiumHD objects each corresponding to one sample. The provided spacerangerOut folder should contain a binned\_outputs folder where multiple bin\_size subfolders are present, e.g., square\_002um.

#### Usage

```
TENxVisiumHD(
  resources,
  spatialResource,
  spacerangerOut,
  sample_id = "sample01",
  processing = c("filtered", "raw"),
  format = c("mtx", "h5"),
  images = c("lowres", "hires", "detected", "aligned_fiducials"),
  bin_size = c("008", "016", "002"),
  jsonFile = .SCALE_JSON_FILE,
  tissuePattern = "tissue_positions\\.parquet",
  spatialCoordsNames = c("pxl_col_in_fullres", "pxl_row_in_fullres"),
  ...
)
```

## S4 method for signature 'TENxVisiumHD,ANY,ANY'
import(con, format, text, ...)

resources	A TENxFileList object or a file path to the tarball containing the matrix / assay data resources.
spatialResource	
	A TENxSpatialList object or a file path to the tarball containing the spatial data.
spacerangerOut	character(1) A single string specifying the path to the sample directory of spaceranger count. The directory must contain the (processing)_feature_bc_matrix and spatial sub directories in addition to the outs folder.
sample_id	character(1) A single string specifying the sample ID.
processing	character(1) A single string indicating the processing folder available e.g., "filtered_feature_barcode_matrix" in the spacerangerOut folder. It can be ei- ther "filtered" or "raw" (default "filtered"). Only used when spacerangerOut is specified.

format	The format of the output. If missing and con is a file name, the format is derived from the file extension. This argument is unnecessary when con is a derivative of BiocFile.
images	character() A vector specifying the images to be imported; can be one or multiple of "lowres", "hires", "detected", "aligned".
bin_size	character(1) The bin size of the images to import. The default is 008. It corresponds to the directory name square_000um where 000 is the bin value.
jsonFile	character(1) A single string specifying the name of the JSON file containing the scale factors.
tissuePattern	character(1) A single string specifying the pattern to match the tissue positions file.
spatialCoordsNa	mes
	character() A vector of strings specifying the names of the columns in the spatial data containing the spatial coordinates.
•••	In the constructor, additional arguments passed to TENxFileList(); otherwise, not used.
con	The connection from which data is loaded or to which data is saved. If this is a character vector, it is assumed to be a file name and a corresponding file connection is created and then closed after exporting the object. If it is a <b>BiocFile</b> derivative, the data is loaded from or saved to the underlying resource. If missing, the function will return the output as a character vector, rather than writing to a connection.
text	If con is missing, this can be a character vector directly providing the string data to import.

#### Details

Typically, the user will provide a path to a directory containing the output of the spaceranger count command. The spaceranger count command outputs a folder containing the "raw" or "filtered" ()\_feature\_bc\_matrix.

#### Value

A SpatialExperiment object

#### Functions

• import(con = TENxVisiumHD, format = ANY, text = ANY): Import Visium HD data from multiple bin sizes

```
vdir <- system.file(
    "extdata", package = "VisiumIO", mustWork = TRUE
)
## with spacerangerOut folder
TENxVisiumHD(spacerangerOut = vdir, bin_size = "002", images = "lowres")</pre>
```

```
TENxVisiumHD(spacerangerOut = vdir, bin_size = "002", images = "lowres") |>
    import()
## indicate h5 format
TENxVisiumHD(
    spacerangerOut = vdir, bin_size = "002",
    images = "lowres", format = "h5"
)
TENxVisiumHD(
    spacerangerOut = vdir, bin_size = "002",
    images = "lowres", format = "h5"
) |>
    import()
## use resources and spatialResource arguments as file paths
TENxVisiumHD(
    resources = file.path(
        vdir, "binned_outputs", "square_002um",
        "filtered_feature_bc_matrix.h5"
    ),
    spatialResource = file.path(
        vdir, "binned_outputs", "square_002um",
        "spatial"
    ),
    bin_size = "002", processing = "filtered",
    images = "lowres", format = "h5"
) |>
    import()
## provide the spatialResource argument as a TENxFileList
TENxVisiumHD(
    resources = file.path(
        vdir, "binned_outputs", "square_002um",
        "filtered_feature_bc_matrix.h5"
    ),
    spatialResource = TENxFileList(
        file.path(
            vdir, "binned_outputs", "square_002um",
            "spatial"
        )
   ),
    bin_size = "002", images = "lowres", format = "h5"
) |>
    import()
```

TENxVisiumList-class A class to represent and import multiple Visium samples

#### TENx VisiumList-class

## Description

This class contains a SimpleList of TENxVisium objects each corresponding to one sample.

## Usage

```
TENxVisiumList(
  sampleFolders,
  sample_ids,
  processing = c("filtered", "raw"),
  images = c("lowres", "hires", "detected", "aligned"),
  format = c("mtx", "h5"),
  jsonFile = .SCALE_JSON_FILE,
  tissuePattern = "tissue_positions.*\\.csv",
  spatialCoordsNames = c("pxl_col_in_fullres", "pxl_row_in_fullres"),
  ...
)
```

## S4 method for signature 'TENxVisiumList,ANY,ANY'
import(con, format, text, ...)

sampleFolders	character() A vector of strings specifying the directories containing the output of the spaceranger count command.	
sample_ids	character() A vector of strings specifying the sample IDs. If not provided, the sample IDs will be the names of the sampleFolders. Therefore, the sample_ids must be the same length as sampleFolders.	
processing	character(1) A single string indicating the processing folder available e.g., "filtered_feature_barcode_matrix" in the spacerangerOut folder. It can be either "filtered" or "raw" (default "filtered"). Only used when spacerangerOut is specified.	
images	character() A vector specifying the images to be imported; can be one or multiple of "lowres", "hires", "detected", "aligned".	
format	The format of the output. If missing and con is a file name, the format is derived from the file extension. This argument is unnecessary when con is a derivative of BiocFile.	
jsonFile	character(1) A single string specifying the name of the JSON file containing the scale factors.	
tissuePattern	character(1) A single string specifying the pattern to match the tissue positions file.	
spatialCoordsNames		
	character() A vector of strings specifying the names of the columns in the spatial data containing the spatial coordinates.	
	In the constructor, additional arguments passed to TENxFileList(); otherwise, not used.	

con	The connection from which data is loaded or to which data is saved. If this is a character vector, it is assumed to be a file name and a corresponding file connection is created and then closed after exporting the object. If it is a <b>BiocFile</b> derivative, the data is loaded from or saved to the underlying resource. If missing, the function will return the output as a character vector, rather than writing to a connection.
text	If con is missing, this can be a character vector directly providing the string data to import.

## Details

Typically, the user will provide a path to a directory containing the output of the spaceranger count command. The spaceranger count command outputs a folder containing the "raw" or "filtered" ()\_feature\_bc\_matrix.

## Value

A SpatialExperiment object

#### Functions

import(con = TENxVisiumList, format = ANY, text = ANY): Import multiple Visium samples

#### See Also

https://support.10xgenomics.com/spatial-gene-expression/software/pipelines/latest/ output/overview

```
sample_dirs <- list.dirs(
    system.file(
        file.path("extdata", "10xVisium"),
        package = "VisiumIO"
    ),
    recursive = FALSE, full.names = TRUE
)
tvl <- TENxVisiumList(
    sampleFolders = sample_dirs,
    sample_ids = c("sample01", "sample02"),
    processing = "raw",
    images = "lowres",
    format = "mtx"
)
import(tvl)</pre>
```

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