

# Package ‘spatialFDA’

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**Title** A Tool for Spatial Multi-sample Comparisons

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**BugReports** <https://github.com/mjemons/spatialFDA/issues>

**Description** spatialFDA is a package to calculate spatial statistics metrics.

The package takes a SpatialExperiment object and calculates spatial statistics metrics using the package spatstat.

Then it compares the resulting functions across samples/conditions using functional additive models as implemented in the package refund.

Furthermore, it provides exploratory visualisations using functional principal component analysis, as well implemented in refund.

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---

.dfToppp	<i>Convert SpatialExperiment object to ppp object</i>
----------	---

---

### Description

Convert SpatialExperiment object to ppp object

### Usage

```
.dfToppp(df, marks = NULL, continuous = FALSE, window = NULL)
```

### Arguments

df	A dataframe with the x and y coordinates from the corresponding SpatialExperiment and the ColData
marks	A vector of marks to be associated with the points, has to be either named 'cell_type' if you want to compare discrete celltypes or else continuous gene expression measurements are assumed as marks.

continuous      A boolean indicating whether the marks are continuous defaults to FALSE  
window            An observation window of the point pattern of class owin.

**Value**

A ppp object for use with spatstat functions

**Examples**

```
spe <- imcdatasets::Damond_2019_Pancreas("spe", full_dataset = FALSE)
speSub <- subset(spe, , image_number == "138")
dfSub <- .speToDf(speSub)
pp <- .dfTopp(dfSub, marks = "cell_type")
```

---

.extractMetric	<i>Compute a spatial metric on a SpatialExperiment object</i>
----------------	---

---

**Description**

A function that takes a SpatialExperiment object and computes a spatial statistics function as implemented in spatstat. The output is a spatstat object.

**Usage**

```
.extractMetric(  
  df,  
  selection,  
  fun,  
  marks = NULL,  
  rSeq = NULL,  
  by = NULL,  
  continuous = FALSE,  
  window = NULL,  
  ...  
)
```

**Arguments**

df                    A dataframe with the x and y coordinates from the corresponding SpatialExperiment and the colData  
selection            the mark(s) you want to compare  
fun                   the spatstat function to compute on the point pattern object  
marks                the marks to consider e.g. cell types  
rSeq                  the range of r values to compute the function over  
by                    the spe colData variable(s) to add to the meta data

continuous      A boolean indicating whether the marks are continuous defaults to FALSE  
 window          a observation window for the point pattern of class owin.  
 ...              Other parameters passed to spatstat.explore functions

**Value**

a spatstat metric object with the fov number, the number of points and the centroid of the image

**Examples**

```
spe <- imcdatasets::Damond_2019_Pancreas("spe", full_dataset = FALSE)
speSub <- subset(spe, , image_number == "138")
dfSub <- .speToDf(speSub)
metricRes <- .extractMetric(dfSub, c("alpha", "beta"),
  fun = "Gcross",
  marks = "cell_type", rSeq = seq(0, 1000, length.out = 100),
  by = c("patient_stage", "patient_id", "image_number")
)
```

---

.speToDf

*Transform a SpatialExperiment into a dataframe*

---

**Description**

Transform a SpatialExperiment into a dataframe

**Usage**

```
.speToDf(spe)
```

**Arguments**

spe              A SpatialExperiment object subset to a single image

**Value**

A dataframe with the x and y coordinates from the corresponding SpatialExperiment and the Col-Data

**Examples**

```
spe <- imcdatasets::Damond_2019_Pancreas("spe", full_dataset = FALSE)
speSub <- subset(spe, , image_number == "138")
dfSub <- .speToDf(speSub)
```

---

calcCrossMetricPerFov *Calculate cross spatial metrics for all combinations per FOV*

---

### Description

A function that takes a `SpatialExperiment` object as input and calculates a cross spatial metric as implemented by `spatstat` per field of view for all combinations provided by the user.

### Usage

```
calcCrossMetricPerFov(
  spe,
  selection,
  subsetby = NULL,
  fun,
  marks = NULL,
  rSeq = NULL,
  by = NULL,
  ncores = 1,
  continuous = FALSE,
  assay = "exprs",
  ...
)
```

### Arguments

<code>spe</code>	a <code>SpatialExperiment</code> object
<code>selection</code>	the mark(s) you want to compare
<code>subsetby</code>	the <code>spe colData</code> variable to subset the data by
<code>fun</code>	the <code>spatstat</code> function to compute on the point pattern object
<code>marks</code>	the marks to consider e.g. cell types
<code>rSeq</code>	the range of <code>r</code> values to compute the function over
<code>by</code>	the <code>spe colData</code> variable(s) to add to the meta data
<code>ncores</code>	the number of cores to use for parallel processing, default = 1
<code>continuous</code>	A boolean indicating whether the marks are continuous defaults to <code>FALSE</code>
<code>assay</code>	the assay which is used if <code>continuous = TRUE</code>
<code>...</code>	Other parameters passed to <code>spatstat.explore</code> functions

### Value

a dataframe of the `spatstat` metric objects with the radius `r`, the theoretical value of a Poisson process, the different border corrections the `fov` number, the number of points and the centroid of the image

**Examples**

```
spe <- imcdatasets::Damond_2019_Pancreas("spe", full_dataset = FALSE)
metricRes <- calcCrossMetricPerFov(spe, c("alpha", "beta"),
  subsetby = "image_number", fun = "Gcross", marks = "cell_type",
  rSeq = seq(0, 50, length.out = 50), by = c(
    "patient_stage", "patient_id",
    "image_number"
  ),
  ncores = 1
)
```

---

calcMetricPerFov	<i>Calculate a spatial metric on a SpatialExperiment object per field of view</i>
------------------	---

---

**Description**

A function that takes a `SpatialExperiment` object as input and calculates a spatial metric as implemented by `spatstat` per field of view.

**Usage**

```
calcMetricPerFov(
  spe,
  selection,
  subsetby,
  fun,
  marks = NULL,
  rSeq = NULL,
  by = NULL,
  continuous = FALSE,
  assay = "exprs",
  ncores = 1,
  ...
)
```

**Arguments**

spe	a <code>SpatialExperiment</code> object
selection	the mark(s) you want to compare
subsetby	the <code>spe colData</code> variable to subset the data by. This variable has to be provided, even if there is only one sample.
fun	the <code>spatstat</code> function to compute on the point pattern object
marks	the marks to consider e.g. cell types
rSeq	the range of r values to compute the function over

by	the spe colData variable(s) to add to the meta data
continuous	A boolean indicating whether the marks are continuous defaults to FALSE
assay	the assay which is used if continuous = TRUE
ncores	the number of cores to use for parallel processing, default = 1
...	Other parameters passed to spatstat.explore functions

**Value**

a dataframe of the spatstat metric objects with the radius r, the theoretical value of a Poisson process, the different border corrections the fov number, the number of points and the centroid of the image

**Examples**

```
spe <- imcdatasets::Damond_2019_Pancreas("spe", full_dataset = FALSE)
metricRes <- calcMetricPerFov(spe, c("alpha", "beta"),
  subsetby = "image_number", fun = "Gcross", marks = "cell_type",
  rSeq = seq(0, 50, length.out = 50), by = c(
    "patient_stage", "patient_id",
    "image_number"
  ),
  ncores = 1
)
```

functionalGam

*General additive model with functional response***Description**

A function that takes the output of a metric calculation as done by calcMetricPerFov. The data has to be prepared into the correct format for the functional analysis by the prepData function. The output is a pffr object as implemented by refund.

**Usage**

```
functionalGam(data, x, designmat, weights, formula, family = "gaussian", ...)
```

**Arguments**

data	a dataframe with the following columns: Y = functional response; sample_id = sample ID; image_id = image ID;
x	the x-axis values of the functional response
designmat	a design matrix as defined by model.matrix()
weights	weights as the number of points per image. These weights are normalised by the mean of the weights in the fitting process

formula	the formula for the model. The colnames of the designmatrix have to correspond to the variables in the formula.
family	the distributional family as implemented in <code>family.mgcv</code> . For fast computation the default is set to <code>gaussian</code> . If the covariance scales e.g. as a function of the domain, this estimation can be improved with <code>gaulss</code> - for more information see <code>family.mgcv</code> .
...	Other parameters passed to <code>pffr</code>

**Value**

a fitted `pffr` object which inherits from `gam`

**Examples**

```
# load the pancreas dataset
library("tidyr")
library("dplyr")
spe <- imcdatasets::Damond_2019_Pancreas("spe", full_dataset = FALSE)
# calculate the Gcross metric for alpha and beta cells
metricRes <- calcMetricPerFov(spe, c("alpha", "beta"),
  subsetby = "image_number", fun = "Gcross",
  marks = "cell_type", rSeq = seq(0, 50, length.out = 50),
  c("patient_stage", "patient_id", "image_number"), ncores = 1
)
metricRes$ID <- paste0(
  metricRes$patient_stage, "|", metricRes$patient_id,
  "|", metricRes$image_number
)
# prepare data for FDA
dat <- prepData(metricRes, "r", "rs")

# drop rows with NA
dat <- dat |> drop_na()

# create meta info of the IDs
splitData <- strsplit(dat$ID, "|", fixed = TRUE)
dat$condition <- factor(sapply(splitData, function(x) x[1]))
dat$patient_id <- factor(sapply(splitData, function(x) x[2]))
dat$image_id <- factor(sapply(splitData, function(x) x[3]))
# create a designmatrix
condition <- dat$condition
# relevel the condition - can set explicit contrasts here
condition <- relevel(condition, "Non-diabetic")
designmat <- model.matrix(~condition)
# colnames don't work with the '-' sign
colnames(designmat) <- c(
  "Intercept", "conditionLong_duration",
  "conditionOnset"
)
# fit the model
mdl <- functionalGam(
  data = dat, x = metricRes$r |> unique(),
```



```

    designmat = designmat, weights = dat$points,
    formula = formula(Y ~ conditionLong_duration +
      conditionOnset + s(patient_id, bs = "re"))
  )
  summary mdl)

```

---

functionalPCA

*Functional Principal Component Analysis*


---

### Description

A function that takes as input the output of `calcMetricPerFov` which has to be converted into the correct format by `prepData`. The output is a list with the `fpc`.`face` output from `refund`.

### Usage

```
functionalPCA(data, r, ...)
```

### Arguments

<code>data</code>	a data object for functional data analysis containing at least the functional response $Y$ .
<code>r</code>	the functional domain
<code>...</code>	Other parameters passed to <code>fpc</code> . <code>sc</code> functions

### Value

a list with components of `fpc`.`sc`

### Examples

```

# load the pancreas dataset
library("tidyr")
library("stringr")
library("dplyr")
spe <- imcdatasets::Damond_2019_Pancreas("spe", full_dataset = FALSE)
# calculate the Gcross metric for alpha and beta cells
metricRes <- calcMetricPerFov(spe, c("alpha", "beta"),
  subsetby = "image_number", fun = "Gcross",
  marks = "cell_type", rSeq = seq(0, 50, length.out = 50),
  c("patient_stage", "patient_id", "image_number"), ncores = 1
)
metricRes$ID <- paste0(
  metricRes$patient_stage, "|", metricRes$patient_id,
  "|", metricRes$image_number
)
# prepare data for FDA
dat <- prepData(metricRes, "r", "rs")

```

```

# drop rows with NA
dat <- dat |> drop_na()
# create meta info of the IDs
splitData <- str_split(dat$ID, "x")
dat$condition <- factor(sapply(splitData, function(x) x[1]))
dat$patient_id <- factor(sapply(splitData, function(x) x[2]))
dat$image_id <- factor(sapply(splitData, function(x) x[3]))
# calculate fPCA
mdl <- functionalPCA(
  data = dat, r = metricRes$r |> unique()
)

```

---

plotCrossFOV

*Creates a  $n \times n$  plot of the cross metrics per sample*


---

### Description

Helper function for plotCrossMetricPerFov. It applies plotMetricPerFov to all n marks defined in the variable selection. This gives an  $n \times n$  plot of all marks.

### Usage

```

plotCrossFOV(
  subFov,
  theo,
  correction,
  x,
  imageId,
  ID = NULL,
  ncol = NULL,
  nrow = NULL,
  legend.position = "none",
  ...
)

```

### Arguments

subFov	a subset of the dataframe to the respective fov
theo	logical; if the theoretical line should be plotted
correction	the border correction to plot
x	the x-axis variable to plot
imageId	the ID of the image/fov
ID	the (optional) ID for plotting combinations
ncol	the number of columns for the facet wrap
nrow	the number of rows for the facet wrap
legend.position	the position of the legend of the plot
...	Other parameters passed to ggplot2 functions

**Value**

a ggplot object

---

plotCrossMetricPerFov *Plot a cross type spatial metric per field of view*

---

**Description**

This function plots the cross function between two marks output from `calcMetricPerFov`. It wraps around helper function and applies this function to all samples.

**Usage**

```
plotCrossMetricPerFov(
  metricDf,
  theo = NULL,
  correction = NULL,
  x = NULL,
  imageId = NULL,
  ID = NULL,
  nrow = NULL,
  ncol = NULL,
  legend.position = "none",
  ...
)
```

**Arguments**

<code>metricDf</code>	the metric dataframe as calculated by <code>calcMetricPerFov</code>
<code>theo</code>	logical; if the theoretical line should be plotted
<code>correction</code>	the border correction to plot
<code>x</code>	the x-axis variable to plot
<code>imageId</code>	the ID of the image/fov
<code>ID</code>	the (optional) ID for plotting combinations
<code>nrow</code>	the number of rows for the facet wrap
<code>ncol</code>	the number of columns for the facet wrap
<code>legend.position</code>	the position of the legend of the plot
<code>...</code>	Other parameters passed to <code>ggplot2</code> functions

**Value**

a ggplot object

**Examples**

```
spe <- imcdatasets::Damond_2019_Pancreas("spe", full_dataset = FALSE)
metricRes <- calcCrossMetricPerFov(spe, c("alpha", "beta"),
  subsetby = "image_number", fun = "Gcross", marks = "cell_type",
  rSeq = seq(0, 50, length.out = 50), by = c(
    "patient_stage", "patient_id",
    "image_number"
  ),
  ncores = 1
)

metricRes$ID <- paste0(
  metricRes$patient_stage, "|", metricRes$patient_id
)

metricRes <- subset(metricRes, image_number %in% c(138, 139, 140))
p <- plotCrossMetricPerFov(metricRes,
  theo = TRUE, correction = "rs",
  x = "r", imageId = "image_number", ID = "ID"
)
print(p)
```

---

plotFbPlot

*Functional boxplot of spatstat curves*


---

**Description**

This function creates a functional boxplot of the spatial statistics curves. It creates one functional boxplot per aggregation category, e.g. condition.

**Usage**

```
plotFbPlot(metricDf, x, y, aggregateBy)
```

**Arguments**

metricDf	the metric dataframe as calculated by calcMetricPerFov
x	the name of the x-axis of the spatial metric
y	the name of the y-axis of the spatial metric
aggregateBy	the criterion by which to aggregate the curves into a functional boxplot. Can be e.g. the condition of the different samples.

**Value**

a list of base R plots

**Examples**

```
spe <- imcdatasets::Damond_2019_Pancreas("spe", full_dataset = FALSE)
metricRes <- calcMetricPerFov(spe, c("alpha", "beta"),
  subsetby = "image_number", fun = "Gcross", marks = "cell_type",
  rSeq = seq(0, 50, length.out = 50), by = c(
    "patient_stage", "patient_id",
    "image_number"
  ),
  ncores = 1
)
# create a unique ID for the data preparation
metricRes$ID <- paste0(
  metricRes$patient_stage, "|", metricRes$patient_id,
  "|", metricRes$image_number
)

plotFbPlot(metricRes, 'r', 'rs', 'patient_stage')
```

---

plotFpca

*Plot a biplot from an fPCA analysis*


---

**Description**

A function that takes the output from the functionalPCA function and returns a ggplot object of the first two dimensions of the PCA as biplot.

**Usage**

```
plotFpca(data, res, colourby = NULL, labelby = NULL)
```

**Arguments**

data	a data object for functional data analysis containing at least the functional response $Y$ .
res	the output from the fPCA calculation
colourby	the variable by which to colour the PCA plot by
labelby	the variable by which to label the PCA plot by

**Value**

a list with components of fpca.face

**Examples**

```

# load the pancreas dataset
library("tidyr")
library("stringr")
library("dplyr")
spe <- imcdatasets::Damond_2019_Pancreas("spe", full_dataset = FALSE)
# calculate the Gcross metric for alpha and beta cells
metricRes <- calcMetricPerFov(spe, c("alpha", "beta"),
  subsetby = "image_number", fun = "Gcross",
  marks = "cell_type", rSeq = seq(0, 50, length.out = 50),
  c("patient_stage", "patient_id", "image_number"), ncores = 1
)
metricRes$ID <- paste0(
  metricRes$patient_stage, "|", metricRes$patient_id,
  "|", metricRes$image_number
)

# prepare data for FDA
dat <- prepData(metricRes, "r", "rs")

# drop rows with NA
dat <- dat |> drop_na()
# create meta info of the IDs
splitData <- str_split(dat$ID, "|")
dat$condition <- factor(sapply(splitData, function(x) x[1]))
dat$patient_id <- factor(sapply(splitData, function(x) x[2]))
dat$image_id <- factor(sapply(splitData, function(x) x[3]))
# calculate fPCA
mdl <- functionalPCA(
  data = dat, r = metricRes$r |> unique()
)
p <- plotFpca(
  data = dat, res = mdl, colourby = "condition",
  labelby = "patient_id"
)
print(p)

```

---

plotMdl

*Plot a pffr model object*


---

**Description**

A function that takes a pffr object as calculated in functionalGam and plots the functional coefficients. The functions are centered such that their expected value is zero. Therefore, the scalar intercept has to be added to the output with the argument shift in order to plot the coefficients in their original range.

**Usage**

```
plotMdl(mdl, predictor, shift = NULL)
```

**Arguments**

mdl	a pffr model object
predictor	predictor to plot
shift	the value by which to shift the centered functional intercept. this will most often be the constant intercept

**Value**

ggplot object of the functional estimate

**Examples**

```

library("tidyr")
library("stringr")
library("dplyr")
spe <- imcdatasets::Damond_2019_Pancreas("spe", full_dataset = FALSE)
metricRes <- calcMetricPerFov(spe, c("alpha", "beta"),
  subsetby = "image_number", fun = "Gcross", marks = "cell_type",
  rSeq = seq(0, 50, length.out = 50), by = c(
    "patient_stage", "patient_id",
    "image_number"
  ),
  ncores = 1
)
# create a unique ID for each row
metricRes$ID <- paste0(
  metricRes$patient_stage, "x", metricRes$patient_id,
  "x", metricRes$image_number
)

dat <- prepData(metricRes, "r", "rs")

# create meta info of the IDs
splitData <- str_split(dat$ID, "x")
dat$condition <- factor(sapply(splitData, function(x) x[1]))
dat$patient_id <- factor(sapply(splitData, function(x) x[2]))
dat$image_id <- factor(sapply(splitData, function(x) x[3]))
# create a designmatrix
condition <- dat$condition
# relevel the condition - can set explicit contrasts here
condition <- relevel(condition, "Non-diabetic")
designmat <- model.matrix(~condition)
# colnames don't work with the '-' sign
colnames(designmat) <- c(
  "Intercept", "conditionLong_duration",
  "conditionOnset"
)
# fit the model
mdl <- functionalGam(
  data = dat, x = metricRes$r |> unique(),
  designmat = designmat, weights = dat$npoints,

```

```

    formula = formula(Y ~ conditionLong_duration +
                      conditionOnset + s(patient_id, bs = "re"))
  )
  summary mdl
  plotLs <- lapply(colnames(designmat), plotMdl,
                  mdl = mdl,
                  shift = mdl$coefficients[["(Intercept)"]]
  )

```

---

plotMetricPerFov      *Plot a spatial metric per field of view*

---

### Description

A function that plots the output of the function calcMetricPerFov. The plot contains one curve per FOV and makes subplots by samples.

### Usage

```

plotMetricPerFov(
  metricDf,
  theo = FALSE,
  correction = NULL,
  x = NULL,
  imageId = NULL,
  ID = NULL,
  nrow = NULL,
  ncol = NULL,
  legend.position = "none",
  ...
)

```

### Arguments

metricDf	the metric dataframe as calculated by calcMetricPerFov
theo	logical; if the theoretical line should be plotted
correction	the border correction to plot
x	the x-axis variable to plot
imageId	the ID of the image/fov
ID	the (optional) ID for plotting combinations
nrow	the number of rows for the facet wrap
ncol	the number of columns for the facet wrap
legend.position	the position of the legend of the plot
...	Other parameters passed to ggplot2 functions



**Value**

a ggplot object

**Examples**

```
spe <- imcdatasets::Damond_2019_Pancreas("spe", full_dataset = FALSE)
metricRes <- calcMetricPerFov(spe, c("alpha", "beta"),
  subsetby = "image_number", fun = "Gcross", marks = "cell_type",
  rSeq = seq(0, 50, length.out = 50), by = c(
    "patient_stage", "patient_id",
    "image_number"
  ),
  ncores = 1
)
# ceate a unique plotting ID
metricRes$ID <- paste0(
  metricRes$patient_stage, "|", metricRes$patient_id
)

p <- plotMetricPerFov(metricRes,
  correction = "rs", x = "r",
  imageId = "image_number", ID = "ID"
)
print(p)
```

---

```
prepData
```

---

*Prepare data from calcMetricRes to be in the right format for FDA*

---

**Description**

Prepare data from calcMetricRes to be in the right format for FDA

**Usage**

```
prepData(metricRes, x, y)
```

**Arguments**

metricRes	a dataframe as calculated by calcMetricRes - requires the columns ID (unique identifier of each row)
x	the name of the x-axis of the spatial metric
y	the name of the y-axis of the spatial metric

**Value**

returns a list with three entries, the unique ID, the functional response Y and the weights

**Examples**

```
spe <- imcdatasets::Damond_2019_Pancreas("spe", full_dataset = FALSE)
metricRes <- calcMetricPerFov(spe, c("alpha", "beta"),
  subsetby = "image_number", fun = "Gcross", marks = "cell_type",
  rSeq = seq(0, 50, length.out = 50), by = c(
    "patient_stage", "patient_id",
    "image_number"
  ),
  ncores = 1
)

# create a unique ID for each row
metricRes$ID <- paste0(
  metricRes$patient_stage, "|", metricRes$patient_id,
  "|", metricRes$image_number
)
dat <- prepData(metricRes, "r", "rs")
```

---

```
print.fpca
```

```
print the fPCA results
```

---

**Description**

this is a function that prints a summary of the fPCA result of class fpca

**Usage**

```
## S3 method for class 'fpca'
print(x, ...)
```

**Arguments**

```
x          the result of function functionalPCA
...        other parameters passed to base generic function print
```

**Value**

a formatted overview of the fPCA result

**Examples**

```
# load the pancreas dataset
library("tidyr")
library("stringr")
library("dplyr")
spe <- imcdatasets::Damond_2019_Pancreas("spe", full_dataset = FALSE)
# calculate the Gcross metric for alpha and beta cells
metricRes <- calcMetricPerFov(spe, c("alpha", "beta"),
  subsetby = "image_number", fun = "Gcross",
```

```
marks = "cell_type", rSeq = seq(0, 50, length.out = 50),
c("patient_stage", "patient_id", "image_number"), ncores = 1
)
metricRes$ID <- paste0(
  metricRes$patient_stage, "|", metricRes$patient_id,
  "|", metricRes$image_number
)
# prepare data for FDA
dat <- prepData(metricRes, "r", "rs")

# drop rows with NA
dat <- dat |> drop_na()

# create meta info of the IDs
splitData <- strsplit(dat$ID, "|", fixed = TRUE)
dat$condition <- factor(sapply(splitData, function(x) x[1]))
dat$patient_id <- factor(sapply(splitData, function(x) x[2]))
dat$image_id <- factor(sapply(splitData, function(x) x[3]))
# calculate fPCA
mdl <- functionalPCA(
  data = dat, r = metricRes$r |> unique()
)
mdl
```

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