Package: LDAcoop (via r-universe)

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Type Package

Title Analysis of Data from Limiting Dilution Assay (LDA) with or without Cellular Cooperation

Version 0.1.2

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URL https://github.com/ZytoHMGU/LDAcoop

BugReports https://github.com/ZytoHMGU/LDAcoop/issues

Description Cellular cooperation compromises the established method of calculating clonogenic activity from limiting dilution assay (LDA) data. This tool provides functions that enable robust analysis in presence or absence of cellular cooperation. The implemented method incorporates the same cooperativity module to model the non-linearity associated with cellular cooperation as known from the colony formation assay (Brix et al. (2021) <doi:10.1038/s41596-021-00615-0>: ``Analysis of clonogenic growth in vitro." Nature protocols).

License GPL-3

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`LDA_activity` *LDA_activity*

Description

calculation of clonogenic activities from data collected in a limiting dilution assay (LDA) experiment (i.e. cells, wells, positive wells, group).

Usage

```
LDA_activity(x, name = "LDA cells")
```

Arguments

`x`	numeric data.frame or matrix with three columns (cells, wells, positive wells, group (optional))
`name`	optional: experiment name (e.g. name of cell line)

Value

list object with LDA-activities as returned by LDA_activity_single

Examples

```
x <- data.frame("cells" = c(10,50,100,250,10,50,100,250),
                "wells" = rep(25,8),
                "positive" = c(2,5,10,20,1,2,6,11),
                "group" = c(rep("A",4),rep("B",4)))
act <- LDA_activity(x)
```

LDA_activity_single *LDA_activity_single*

Description

calculation of clonogenic activity from data collected by a limiting dilution assay (LDA) experiment (i.e. numbers of: cells seeded, wells, positive wells).

Usage

```
LDA_activity_single(x, name = "cell line a", treat = "no")
```

Arguments

x	numeric data.frame or matrix with three columns (cells, wells, positive wells)
name	optional: experiment name (e.g. name of cell line)
treat	optional: treatment (e.g. irradiation dose in Gy)

Value

list object with estimated activity, 95 84 matrix, fit-object and p-value for cooperativity-test

Examples

```
x <- data.frame("cells" = c(10,50,100,250),
                "wells" = rep(25,4),
                "positive" = c(2,5,10,20))
act <- LDA_activity_single(x)
data(LDAdata)
cell.line <- unique(LDAdata$name)[1]
x <- subset.data.frame(
      LDAdata,
      subset = (name==cell.line) & (Group == 0))
LDA_activity_single(x[,4:6])
```

LDA_plot *LDA_plot*

Description

plot clonogenic activity and survival (at more than one treatment group) for data from limiting dilution assay (LDA) experiments.

Usage

```
LDA_plot(LDA_tab, uncertainty = "act", xlim = NULL, uncertainty.band = FALSE)
```

Arguments

LDA_tab	LDA data.frame ("cells", "wells", "positive", "group", "replicate")
uncertainty	method for uncertainty calculation ("act", "ep")
xlim	setting xlim of clonogenic activity plot
uncertainty.band	plotting of uncertainty bands TRUE/FALSE

Value

none

Examples

```
data(LDAdata)
Z1 <- subset.data.frame(LDAdata,subset = name == unique(LDAdata$name)[1])
LDA_plot(Z1[,c("S-value","# Tested","# Clonal growth","Group","replicate")])
```

LDA_plot_activity *LDA_plot_activity*

Description

generate clonogenic activity estimation plot (frequency of negative wells over the number of cells seeded) for data of limiting dilution assay (LDA) experiments. Input is an data object as returned by the preprocessing function LDA_prepare_plot().

Usage

```
LDA_plot_activity(LDA_obj, xlim = NULL, uncertainty.band = FALSE)
```

Arguments

LDA_obj	list returned from LDA_prepare_plot
xlim	manually setting the xlim
uncertainty.band	plotting uncertainty bands TRUE/FALSE

Value

none

Examples

```
x <- data.frame("cells" = rep(c(10,50,100,250),times = 4),
                "wells" = rep(25,16),
                "positive" = c(2,5,10,20,1,2,6,11,3,4,8,22,1,1,7,12),
                "group" = rep(c(rep("A",4),rep("B",4)),times = 2),
                "replicate" = c(rep(1,8),rep(2,8)))
out <- LDA_prepare_plot(x)
LDA_plot_activity(out[[1]])
data(LDAdata)
Z1 <- subset.data.frame(LDAdata,subset = name == unique(LDAdata$name)[1])
out <- LDA_prepare_plot(Z1[,c("S-value","# Tested","# Clonal growth",
                              "Group","replicate")])
LDA_plot_activity(out[[1]])
```

LDA_plot_SF *LDA_plot_SF*

Description

generate clonogenic survival plot (estimated clonogenic survival over treatment) for data from limiting dilution assay (LDA). Input is an data object as returned by the preprocessing function LDA_prepare_plot().

Usage

```
LDA_plot_SF(LDA_obj)
```

Arguments

LDA_obj	list returned from LDA_prepare_plot

Value

none

Examples

```
x <- data.frame("cells" = rep(c(10,50,100,250),times = 4),
                "wells" = rep(25,16),
                "positive" = c(2,5,10,20,1,2,6,11,3,4,8,22,1,1,7,12),
                "group" = rep(c(rep(0,4),rep(6,4)),times = 2),
                "replicate" = c(rep(1,8),rep(2,8)))
out <- LDA_prepare_plot(x)
LDA_plot_SF(out[[2]])
data(LDAdata)
Z1 <- subset.data.frame(LDAdata,subset = name == unique(LDAdata$name)[1])
out <- LDA_prepare_plot(Z1[,c("S-value","# Tested","# Clonal growth",
                              "Group","replicate")])
LDA_plot_SF(out[[2]])
```

LDA_prepare_plot *LDA_prepare_plot*

Description

analyze limiting dilution assay (LDA) data and collect information for plotting.

Usage

```
LDA_prepare_plot(LDA_tab, uncertainty = "act")
```

Arguments

LDA_tab	LDA data.frame ("cells", "wells", "positive", "group", "replicate")
uncertainty	method for approximation of uncertainties of survival fractions (SF): activity based ("act") or by error propagation ("ep")

Value

none

Examples

```
x <- data.frame("cells" = rep(c(10,50,100,250),times = 4),
                "wells" = rep(25,16),
                "positive" = c(2,5,10,20,1,2,6,11,3,4,8,22,1,1,7,12),
                "group" = rep(c(rep("A",4),rep("B",4)),times = 2),
                "replicate" = c(rep(1,8),rep(2,8)))
LDA_prepare_plot(x)
# data(LDAdata)
# Z1 <- subset.data.frame(LDAdata,subset = name == unique(LDAdata$name)[1])
# LDA_prepare_plot(Z1[,c("S-value","# Tested","# Clonal growth","Group",
#                        "replicate")])
```

LDA_survival *LDA_survival*

Description

calculation of clonogenic survival in a table of data from a limiting dilution assay (LDA) experiment (i.e. cells, wells, positive wells, group).

Usage

```
LDA_survival(x, name = "cell line a")
```

Arguments

x	numeric data.frame or matrix with three columns (cells, wells, positive wells, group)
name	optional: experiment name (e.g. name of cell line)

Value

list object with LDA-activities as returned by LDA_activity_single

Examples

```
x <- data.frame("cells" = c(10,50,100,250,10,50,100,250),
                "wells" = rep(25,8),
                "positive" = c(2,5,10,20,1,2,6,11),
                "group" = c(rep("A",4),rep("B",4)))
act <- LDA_survival(x)
```

LDA_survival_single *LDA_survival_single*

Description

calculate clonogenic survival fraction from LDA_activity objects.

Usage

```
LDA_survival_single(act.0, act.x)
```

Arguments

act.0	reference activity
act.x	activity after treatment

Value

list object with survival fraction, estimated confidence intervals (by error propagation through first order Taylor series approximation and by combination of 84

Examples

```
x.a <- data.frame("cells" = c(10,50,100,250),
                  "wells" = rep(25,4),
                  "positive" = c(2,5,10,20))
x.b <- data.frame("cells" = c(10,50,100,250),
                  "wells" = rep(25,4),
                  "positive" = c(1,2,6,11))
act.a <- LDA_activity_single(x.a)
act.b <- LDA_activity_single(x.b)
sf <- LDA_survival_single(act.0 = act.a,act.x = act.b)
data(LDAdata)
cell.line <- unique(LDAdata$name)[1]
x <- subset.data.frame(LDAdata, subset = (name==cell.line) & (Group < 2))
act <- LDA_activity(x[,c(4:6,3)])
sf <- LDA_survival_single(act.0 = act[[1]],act.x = act[[2]])
```

LDA_table *LDA_table*

Description

show table with activities and clonogenic survival from limiting dilution assay (LDA) data.

Usage

```
LDA_table(x, ref_class = "unknown", uncertainty = "act")
```

Arguments

x	numeric data.frame or matrix with at least three columns (cells, wells, positive wells, group (optional))
ref_class	name of reference class for calculation of SF values
uncertainty	method for calculating the uncertainty bands of survival fractions ("act" (default) for combining the activity confidence intervals; "ep" for error propagation via first order Taylor series expansion.)

Value

table

Examples

```
x <- data.frame("cells" = c(10,50,100,250,10,50,100,250),
                "wells" = rep(25,8),
                "positive" = c(2,5,10,20,1,2,6,11),
                "group" = c(rep("A",4),rep("B",4)))
LDA_table(x,ref_class = "A")
```

LDAdata *LDA (limiting dilution assay) data from a set of cell lines*

Description

LDA data from 11 cell lines, up to 4 biological replicates and up to 6 treatments.

Usage

```
data(LDAdata)
```

Format

data.frame with columns: "name", "replicate", "Group", "S-value", "# Tested", "# Clonal growth"

Examples

```
data(LDAdata)
```

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