Tutorial -

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1 Aim

This tutorial explain how plot growth curves for phytoplankton cultures in different conditions. * Antibiotics treatment : 4 RCC cultures, 8 days, 2 antibiotics, 5 concentrations,

2 Downloads

Install the following software :

- R studio : https://www.rstudio.com/products/rstudio/download/#download
- Download and install the following libraries by running under R studio the following lines

```
install.packages("dplyr")  # To manipulate dataframes
install.packages("tidyr")  # To manipulate dataframes

install.packages("stringr")  # To strings

install.packages("ggplot2")  # for high quality graphics
install.packages("gridExtra")  # for grids

install.packages("plotrix")  # needed for standard error
```

3 Data used

• cultures02.txt contains data obtained by Priscilla Gourvil on antibiotics treatment of RCC strains

4 Tutorial description

4.1 Load the libraries

```
library("ggplot2")
library("gridExtra")
library("plotrix" ) # needed for standard error
library("dplyr")
library("tidyr")
library("stringr")
```

4.2 Antibiotics treatments

4.2.1 Read and reformat the data

Read the data

```
cell<- read.table("cultures 02.txt", header=TRUE, sep="\t", na.strings="NA", dec=".", strip.white=TRUE)
knitr::kable(head(cell))</pre>
```

| RCC | Antibio | Well | Concentration | X1 | X2 | Х3 | X4 | X5 | X6 | X7 |
|----------|---------|------|---------------|-------|-------|-------|------|-------|-------|--------|
| RCC 96 | G 418 | A01 | 0.5 | 35888 | 17043 | 3853 | 593 | 225 | 675 | 591 |
| RCC 96 | G 418 | A02 | 0.5 | 27281 | 20952 | 337 | 450 | 84 | 394 | 562 |
| RCC 96 | G 418 | A03 | 0.5 | 29952 | 19630 | 34846 | 3853 | 21318 | 19996 | 31639 |
| RCC 96 | G 418 | B01 | 1.0 | 33018 | 17268 | 6468 | 337 | 478 | 394 | 253 |
| RCC 96 | G 418 | B02 | 1.0 | 26662 | 20530 | 34902 | 675 | 22555 | 8268 | 14062 |
| RCC 96 | G 418 | B03 | 1.0 | 24946 | 19040 | 49011 | 4725 | 21599 | 26746 | 277217 |

Change from wide format to long format

```
cell<- gather(cell, X1:X7, key = "day", value = "cell_number")
knitr::kable(head(cell))</pre>
```

| RCC | Antibio | Well | Concentration | day | cell_number |
|--------|---------|------|---------------|-----|-------------|
| RCC 96 | G 418 | A01 | 0.5 | X1 | 35888 |
| RCC 96 | G 418 | A02 | 0.5 | X1 | 27281 |
| RCC 96 | G 418 | A03 | 0.5 | X1 | 29952 |
| RCC 96 | G 418 | B01 | 1.0 | X1 | 33018 |
| RCC 96 | G 418 | B02 | 1.0 | X1 | 26662 |
| RCC 96 | G 418 | B03 | 1.0 | X1 | 24946 |
| | | | | | |

Reformat day as numeric

```
cell$day<-as.numeric(str_replace(cell$day, "X", ""))
knitr::kable(head(cell))</pre>
```

| RCC | Antibio | Well | Concentration | day | cell_number |
|--------|---------|------|---------------|-----|-------------|
| RCC 96 | G 418 | A01 | 0.5 | 1 | 35888 |
| RCC 96 | G 418 | A02 | 0.5 | 1 | 27281 |
| RCC 96 | G418 | A03 | 0.5 | 1 | 29952 |

| RCC | Antibio | Well | Concentration | day | cell_number |
|--------|---------|------|---------------|-----|-------------|
| RCC 96 | G 418 | B01 | 1.0 | 1 | 33018 |
| RCC 96 | G418 | B02 | 1.0 | 1 | 26662 |
| RCC 96 | G 418 | B03 | 1.0 | 1 | 24946 |

Reformat concentration as character

```
cell$Concentration<-as.character(cell$Concentration)
knitr::kable(head(cell))</pre>
```

| RCC | Antibio | Well | Concentration | day | cell_number |
|--------|---------|------|---------------|-----|-------------|
| RCC 96 | G 418 | A01 | 0.5 | 1 | 35888 |
| RCC 96 | G 418 | A02 | 0.5 | 1 | 27281 |
| RCC 96 | G 418 | A03 | 0.5 | 1 | 29952 |
| RCC 96 | G 418 | B01 | 1 | 1 | 33018 |
| RCC 96 | G 418 | B02 | 1 | 1 | 26662 |
| RCC 96 | G 418 | B03 | 1 | 1 | 24946 |

Compute mean and SD for each RCC, Antibio, Concentration and day using dplyr

| RCC | Antibio | Concentration | day | cell_mean | $\operatorname{cell_sd}$ | cell_se |
|----------|---------|---------------|-----|------------|---------------------------|-----------|
| RCC 4094 | G 418 | 0.5 | 1 | 188562.333 | 46983.373 | 27125.863 |
| RCC 4094 | G418 | 0.5 | 2 | 100846.333 | 48010.521 | 27718.887 |
| RCC 4094 | G418 | 0.5 | 3 | 4087.667 | 3401.215 | 1963.692 |
| RCC 4094 | G418 | 0.5 | 4 | 42195.333 | 18511.363 | 10687.541 |
| RCC 4094 | G418 | 0.5 | 5 | 8727.667 | 14751.465 | 8516.763 |
| RCC 4094 | G 418 | 0.5 | 6 | 3806.000 | 6105.543 | 3525.037 |

4.2.2 Define graphics options

Define the color, line type and symbol shape

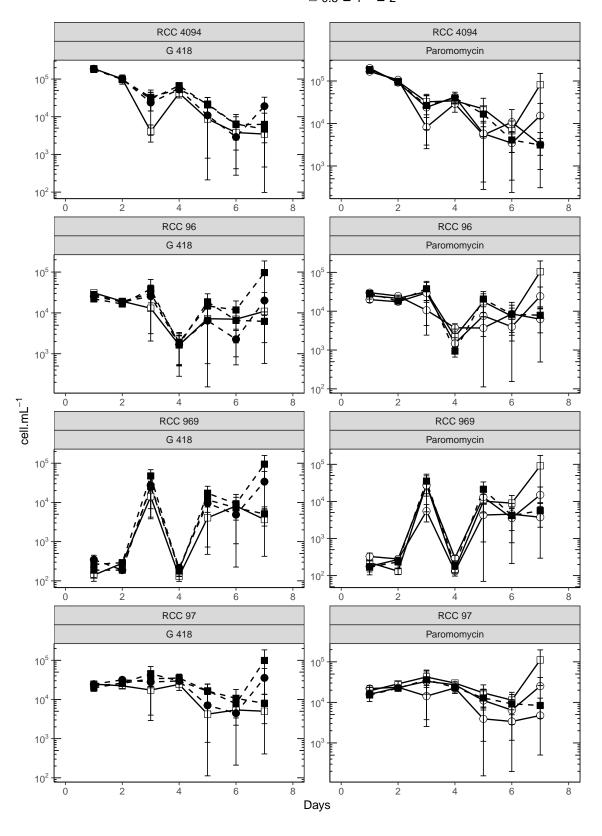
```
 \begin{tabular}{ll} Concentration\_color <-c ("0.2"="white", "0.5"="white", "0.8"="white", "1"="black", "1.5"="black", "2"="black", "0.8"=1, "1"=2, "1.5"=2, "2"=2) \\ Concentration\_shape <-c ("0.2"=21,"0.5"=22, "0.8"=21, "1"=22, "1.5"=21, "2"=22) \\ \end{tabular}
```

Define graphics options

```
scaling_factor=15
cell_label <- expression (paste("cell.",mL^-1))
cell_breaks=c(100,1000,10000,100000,1000000)
x_max=8
x_breaks=c(0, 2,4,6,8)
x_labels=c("0", "2","4","6","8")</pre>
```

4.2.3 Plot the data

```
plot1<- ggplot(cell_1, aes(x=day, y=cell_mean, group = Concentration, xmin=0, xmax=x_max,</pre>
               shape=Concentration, fill=Concentration, linetype=Concentration)) +
              facet_wrap(~ RCC + Antibio, nrow=4, ncol=2, scales="free") +
                geom_line (size=0.8, colour="black") +
              geom point(size = 4) +
              geom_errorbar(aes(ymin=cell_mean-cell_se, ymax=cell_mean+cell_se), width=0.2, linetype=1)
              theme_bw(scaling_factor) +
              theme(panel.border = element_rect(colour = "black"), panel.grid.major = element_blank(),
              axis.line = element_line(colour = "black"),
                  legend.title=element_text(size=scaling_factor), legend.key=element_blank(),
                  axis.title = element_text(size=scaling_factor),
                  legend.text=element_text(size=scaling_factor), legend.key.height = unit(1, "cm"),
                  axis.text = element_text(size=0.8*scaling_factor), panel.background = element_rect(fi
                theme(legend.position = "top", legend.box = "horizontal") +
                labs(x = "Days", y = cell_label ) +
              scale_x_continuous(breaks=x_breaks, labels=x_labels) +
                scale_y_log10(breaks = cell_breaks ,labels = scales::trans_format("log10", scales::math
              annotation_logticks(sides = "lr") +
                scale_fill_manual(values=Concentration_color) +
              scale_shape_manual(values=Concentration_shape) +
              scale_linetype_manual(values=Concentration_linetype)
# Add next line to zoom
# + coord_cartesian(ylim=c(100, 10000000))
plot1
```



```
# Next can be used to save the plot as pdf
# ggsave(file="Fig 1 version 2.0.pdf", plot=plot1, scale=5, width = 7, height = 10, units = "cm", useDi
```