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Microarray Center

Introduction to R:

Statistical Tools for Biologists

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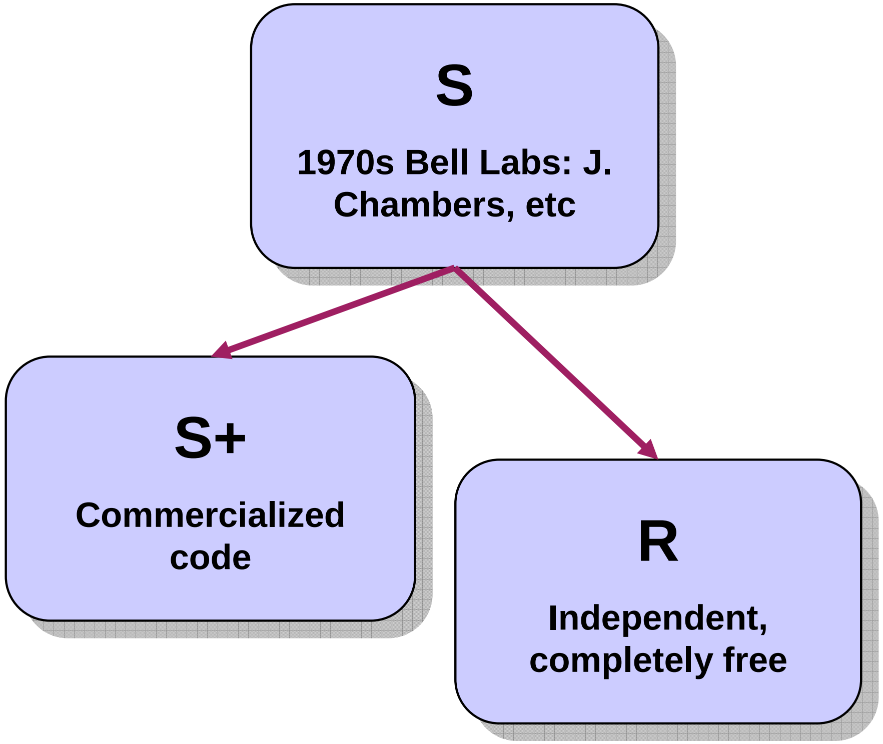
OUTLINE

- **General introduction and information package**
- **Data manipulation and visualization**
  - basic operations
  - import and export
  - visualization
- **Simple statistical analysis**
  - descriptive statistics
  - statistical tests
  - simple ANOVA (to be continued by Thomas)
- **Demonstration of Bioconductor**

1. GENERAL INTRODUCTION

History



S

1970s Bell Labs: J. Chambers, etc

S+

Commercialized code

R

Independent, completely free

R was created by
Ross Ihaka and **Robert Gentleman** at the University of Auckland, New Zealand, and is now developed by the
R Development Core Team.

Positive Features

- Scripting language of high level
- Interactive work with data (similar to MATLAB)
- Works under Windows, Linux, Mac OS
- Free code + verified algorithms
- Fast developing: new version each 2 month
- Extremely wide application: from biology to theoretical physics and computer sciences
- Very good information support
- Fast installation (with some exclusions)

Negative

- Not memory efficient (comparing to C)
- Slower then C/C++
- No advanced built-in GUI development tools

2. INFORMATION PACKAGE

Main Web-page:

cran.r-project.org

cran.r-project.org/manuals.html

cran.r-project.org/web/packages/

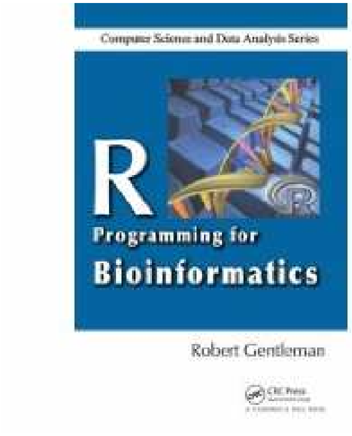
cran.r-project.org/other-docs.html

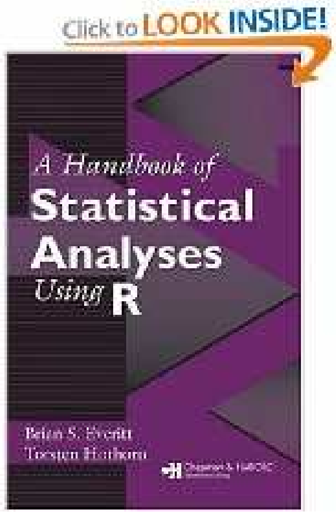
R-Project Seek Engine:

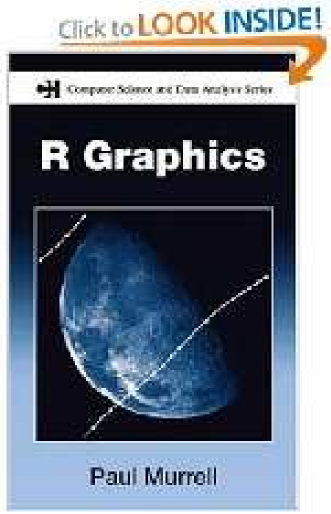
www.rseek.org

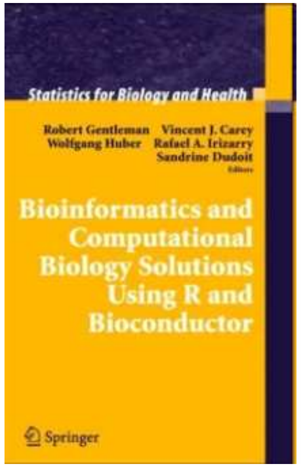
R/Bioconductor

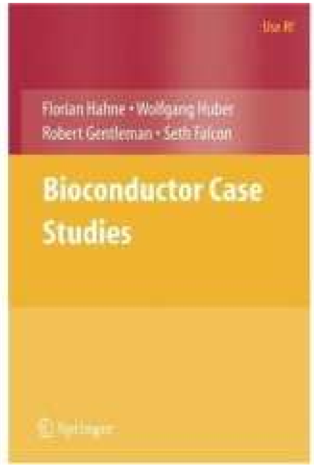
www.bioconductor.org


Computer Science and Data Analysis Series
R Programming for Bioinformatics
Robert Gentleman
CRC Press






Statistics for Biology and Health
Robert Gentleman Vincent J. Carey
Wolfgang Huber Rafael A. Irizarry
Sandrine Dudoit
Editors
Bioinformatics and Computational Biology Solutions Using R and Bioconductor
Springer


Use R!
Florian Hahne • Wolfgang Huber
Robert Gentleman • Seth Falcon
Bioconductor Case Studies
Springer

2. INFORMATION PACKAGE

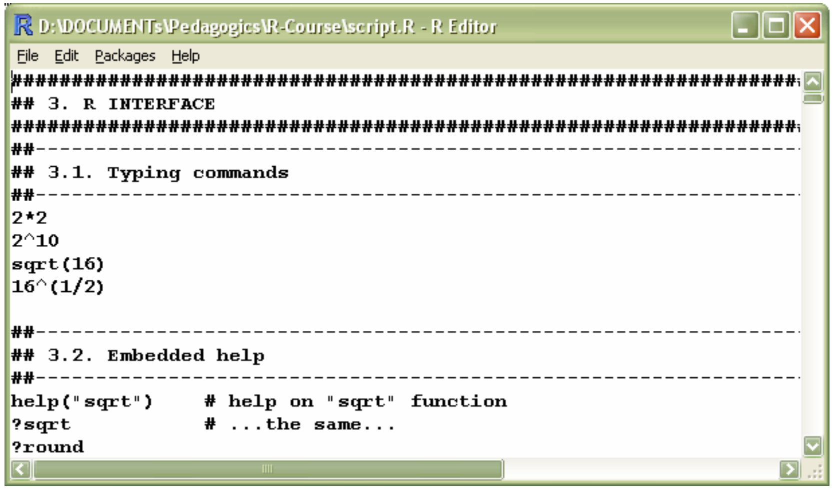
All the materials and data used in the lecture

are available here

```
http://edu.sablab.net/r
```

3. R INTERFACE

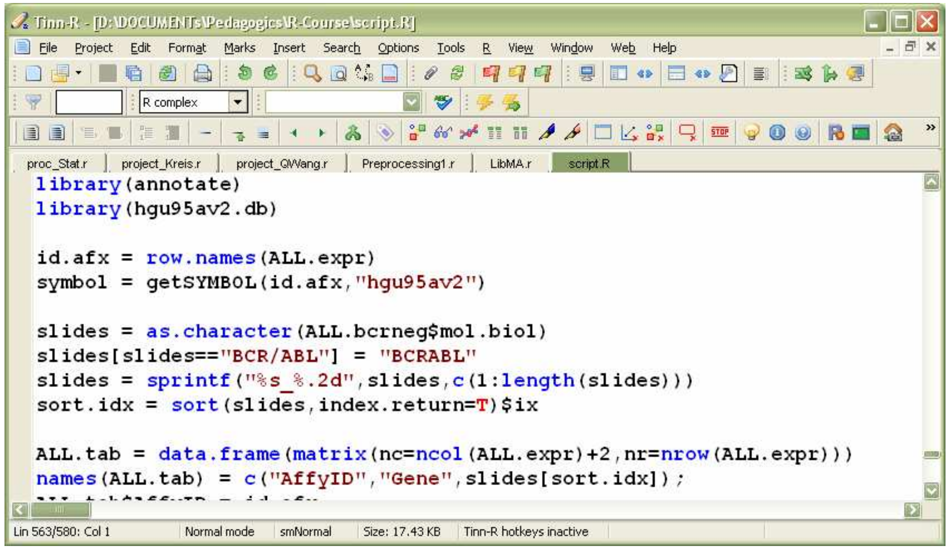
Built-in Script Editor



```
##########################################################################
## 3. R INTERFACE
##########################################################################
##------------------------------------------------------------------------
## 3.1. Typing commands
##------------------------------------------------------------------------
2*2
2^10
sqrt(16)
16^(1/2)

##------------------------------------------------------------------------
## 3.2. Embedded help
##------------------------------------------------------------------------
help("sqrt")     # help on "sqrt" function
?sqrt            # ...the same...
?round
```

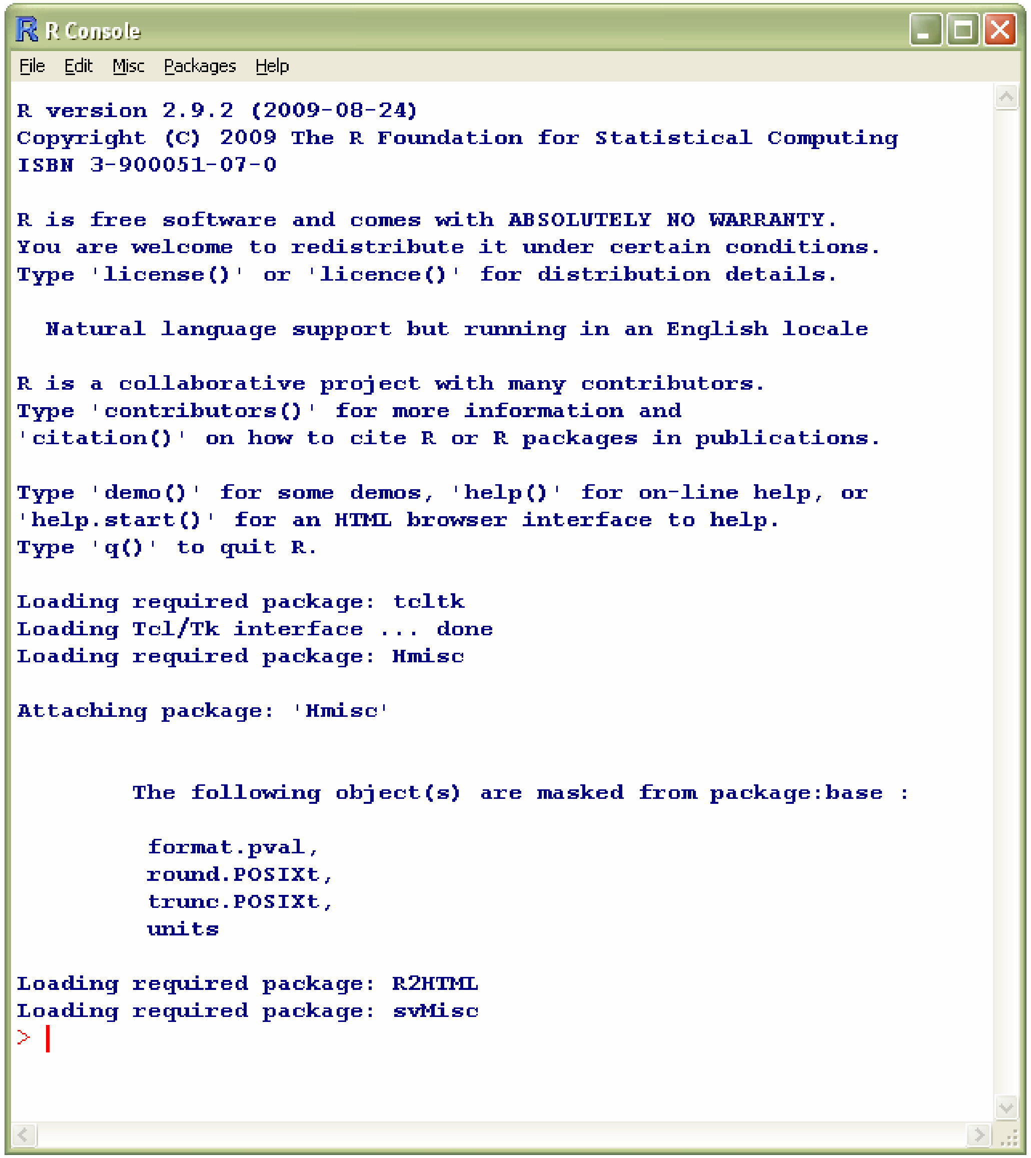
Alternative: Tinn-R

(highly recommended for win-users)



```
library(annotate)
library(hgu95av2.db)

id.afx = row.names(ALL.expr)
symbol = getSYMBOL(id.afx,"hgu95av2")

slides = as.character(ALL.bcrneg$mol.biol)
slides[slides=="BCR/ABL"] = "BCRABL"
slides = sprintf("%s_%.2d",slides,c(1:length(slides)))
sort.idx = sort(slides,index.return=T)$ix

ALL.tab = data.frame(matrix(nc=ncol(ALL.expr)+2,nr=nrow(ALL.expr)))
names(ALL.tab) = c("AffyID","Gene",slides[sort.idx]);
```

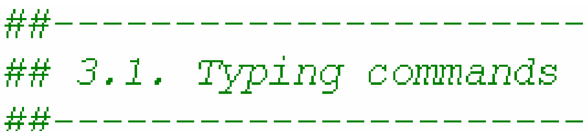
Console



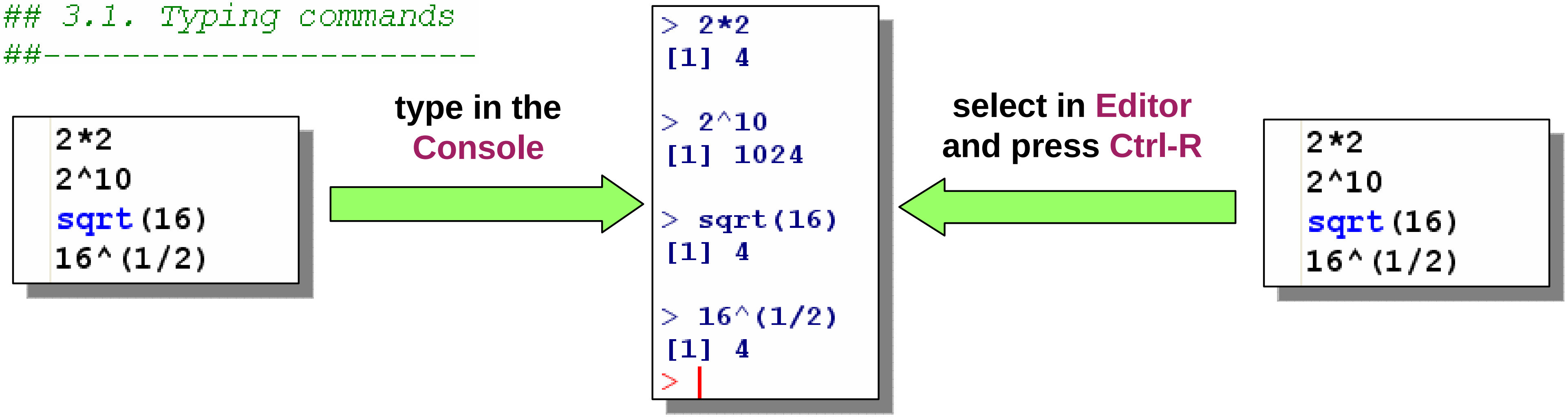
```
R version 2.9.2 (2009-08-24)
Copyright (C) 2009 The R Foundation for Statistical Computing
ISBN 3-900051-07-0

R is free software and comes with ABSOLUTELY NO WARRANTY.
You are welcome to redistribute it under certain conditions.
Type 'license()' or 'licence()' for distribution details.

  Natural language support but running in an English locale

R is a collaborative project with many contributors.
Type 'contributors()' for more information and
'citation()' on how to cite R or R packages in publications.

Type 'demo()' for some demos, 'help()' for on-line help, or
'help.start()' for an HTML browser interface to help.
Type 'q()' to quit R.

Loading required package: tcltk
Loading Tcl/Tk interface ... done
Loading required package: Hmisc

Attaching package: 'Hmisc'


        The following object(s) are masked from package:base :

         format.pval,
         round.POSIXt,
         trunc.POSIXt,
         units

Loading required package: R2HTML
Loading required package: svMisc
> 
```

http://sourceforge.net/projects/tinn-r/

3. R INTERFACE



```
##-----------------------
## 3.1. Typing commands
##-----------------------
```



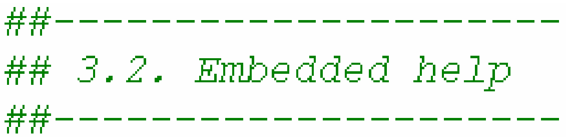
```
2*2
2^10
sqrt(16)
16^(1/2)
```

type in the Console

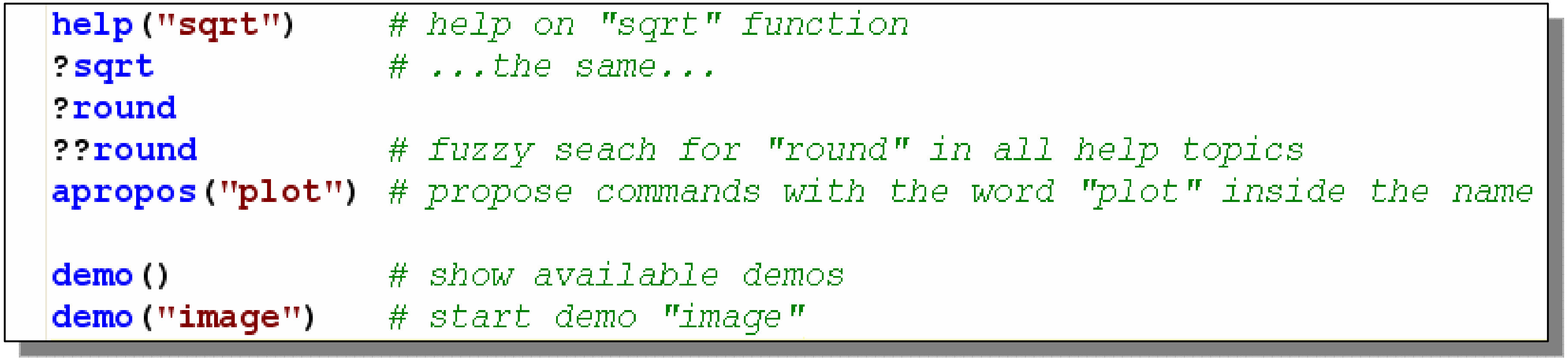
```
> 2*2
[1] 4

> 2^10
[1] 1024

> sqrt(16)
[1] 4

> 16^(1/2)
[1] 4
> 
```

select in Editor and press Ctrl-R

```
2*2
2^10
sqrt(16)
16^(1/2)
```

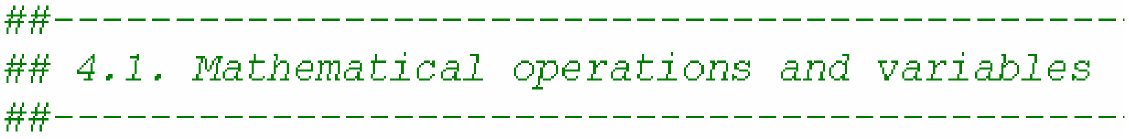


```
##----------------------
## 3.2. Embedded help
##----------------------
```

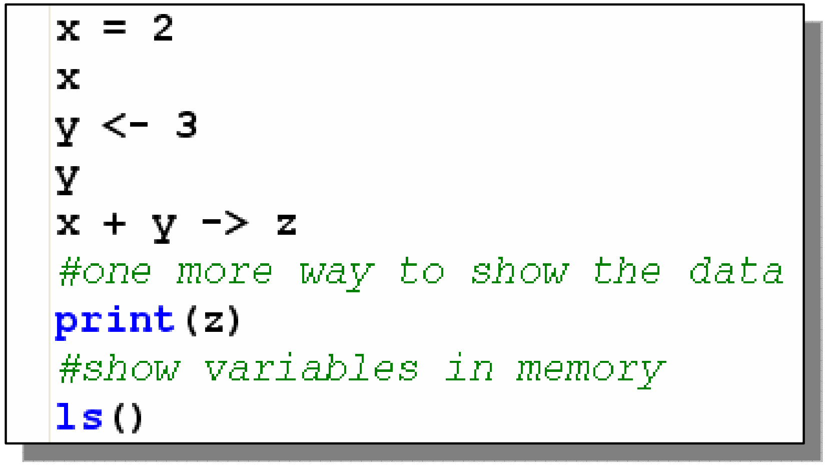


```
help("sqrt")     # help on "sqrt" function
?sqrt            # ...the same...
?round
??round          # fuzzy seach for "round" in all help topics
apropos("plot")  # propose commands with the word "plot" inside the name

demo()           # show available demos
demo("image")    # start demo "image"
```

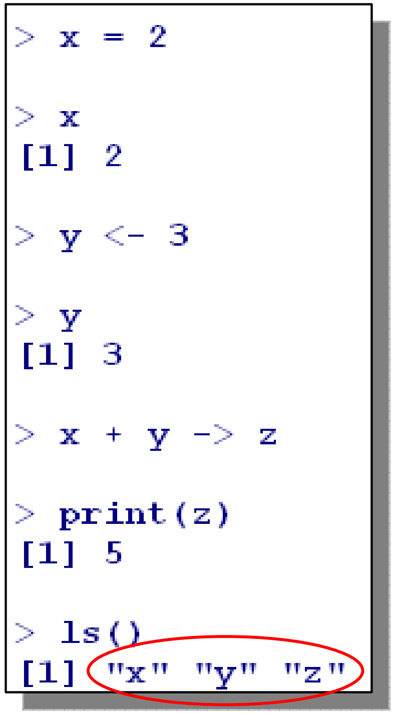
4. BASIC OPERATIONS (4.1)



```
##-------------------------------------------
## 4.1. Mathematical operations and variables
##-------------------------------------------
```

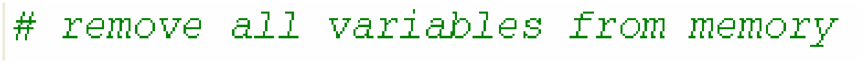


```
x = 2
x
y <- 3
y
x + y -> z
#one more way to show the data
print(z)
#show variables in memory
ls()
```

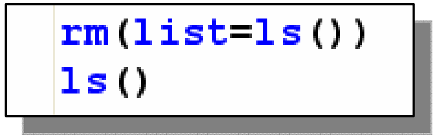


```
> x = 2

> x
[1] 2

> y <- 3

> y
[1] 3

> x + y -> z

> print(z)
[1] 5

> ls()
[1] "x" "y" "z"
```

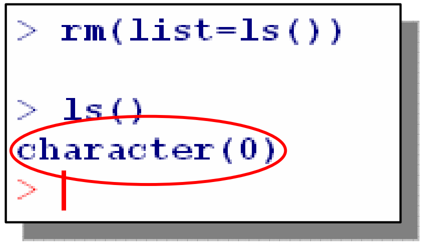
See
“R Reference Card”
for more functions
and details



```
# remove all variables from memory
```



```
rm(list=ls())
ls()
```



```
> rm(list=ls())

> ls()
character(0)
> 
```

4. BASIC OPERATIONS (4.2)

Scalar Data

Numeric

Integer

Double

```
1
3.141593
```

Logical

```
TRUE
FALSE
```

Character

```
"Hello, world!"
```

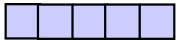
Factor

has a sense to use only in vectors or data frames

```
> answer=factor(c("yes","no"))
> answer
[1] yes no
Levels: no yes
```

Data Containers

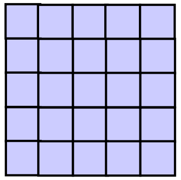
Vector



```
> x
[1] 1 2 3 4 5
```

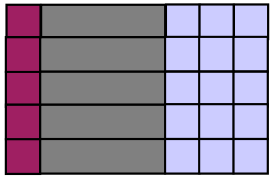
Matrix

Array



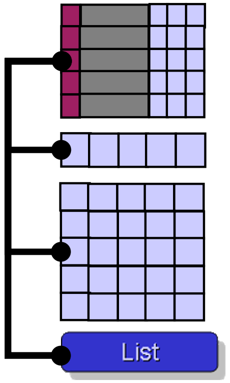
```
> A
     [,1] [,2] [,3]
[1,]    1    3    5
[2,]    2    4    1
```

Data frame

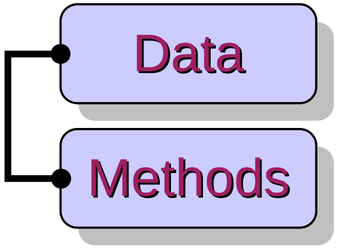


```
   name marks
1  Alex    10
2  Jean     8
3 David     7
```

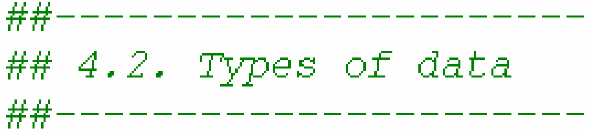
List



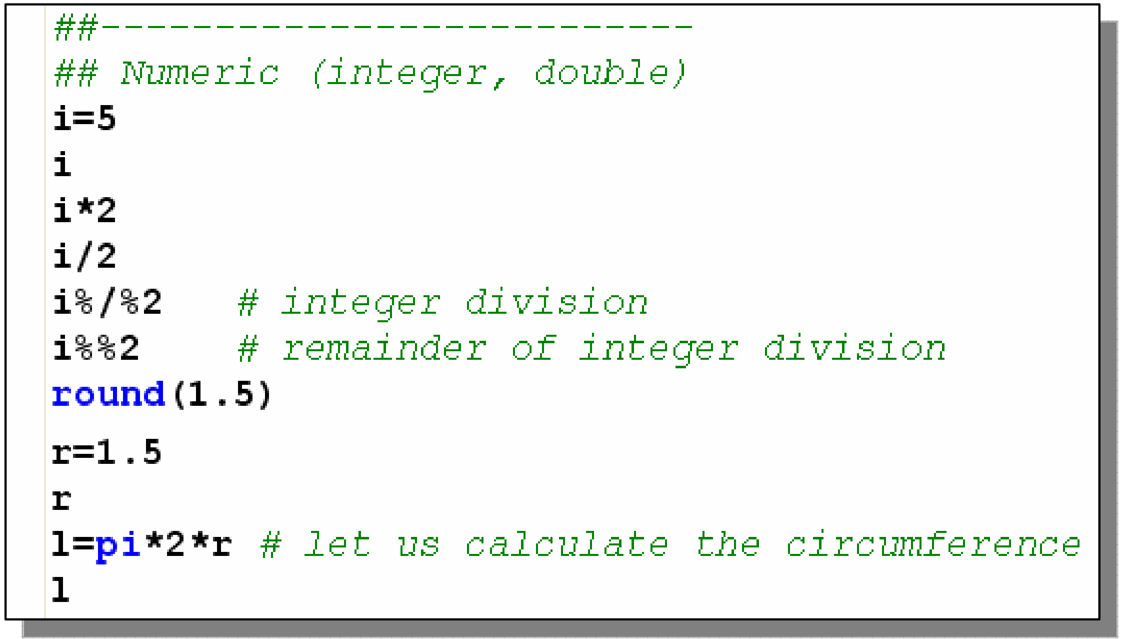
Object of a Class



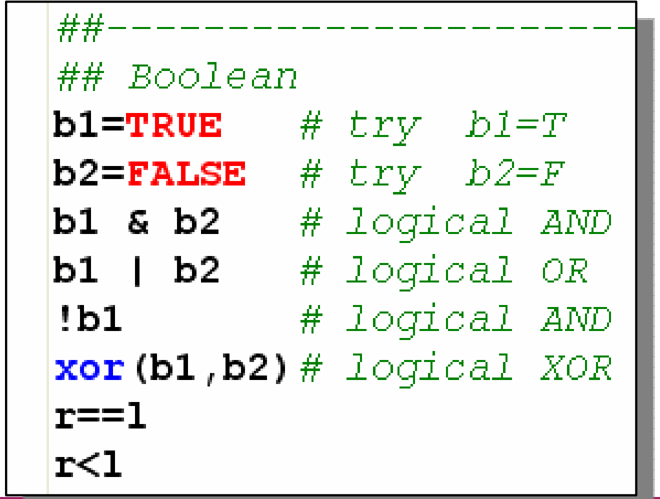
4. BASIC OPERATIONS (4.2)



```
##-----------------------
## 4.2. Types of data
##-----------------------
```

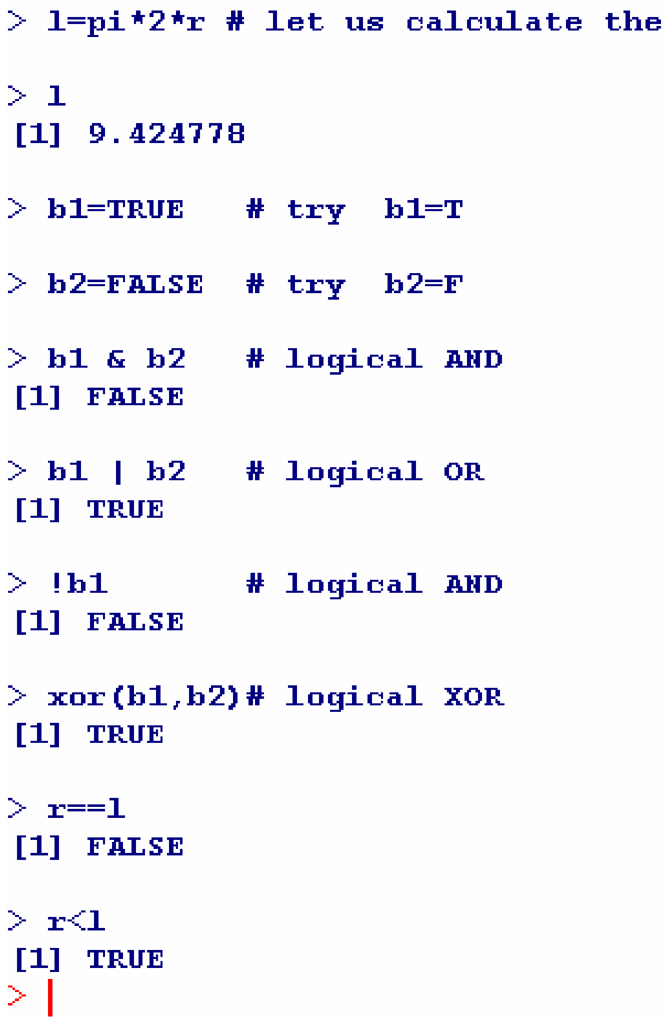


```
##-------------------------
## Numeric (integer, double)
i=5
i
i*2
i/2
i%/%2    # integer division
i%%2     # remainder of integer division
round(1.5)
r=1.5
r
l=pi*2*r # let us calculate the circumference
l
```



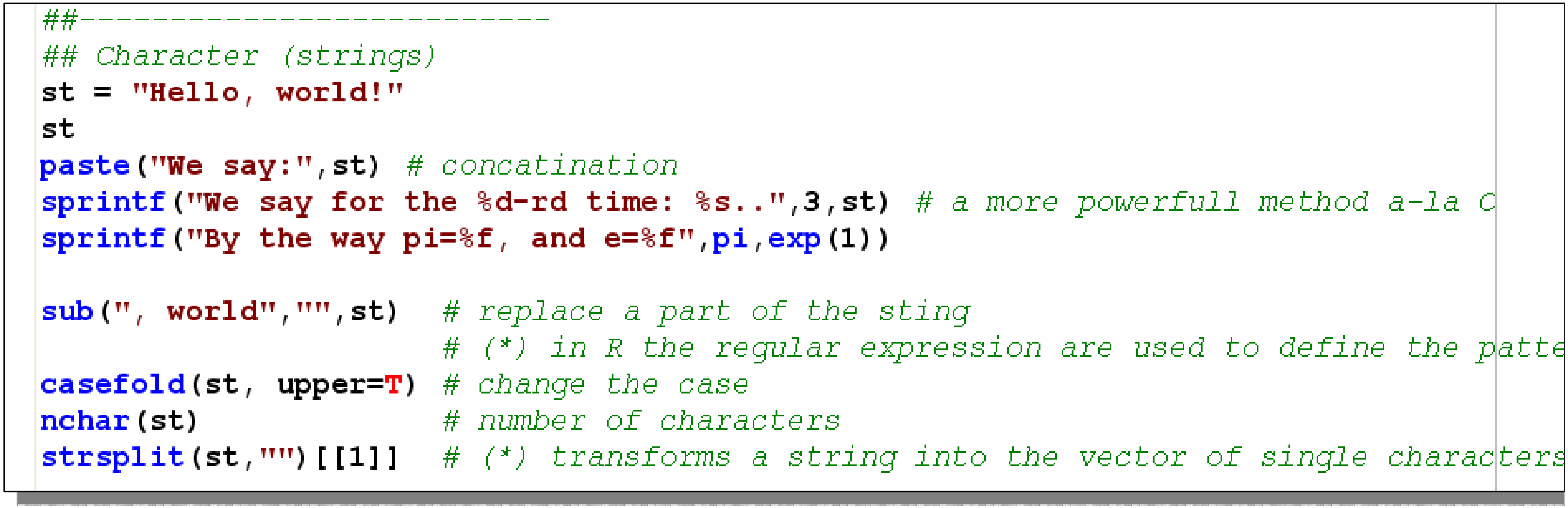
```
##-----------------------
## Boolean
b1=TRUE    # try  b1=T
b2=FALSE   # try  b2=F
b1 & b2    # logical AND
b1 | b2    # logical OR
!b1        # logical AND
xor(b1,b2) # logical XOR
r==l
r<l
```

For integer bitwise operation install and use "bitops" package
bitAnd(), bitOr(), ...

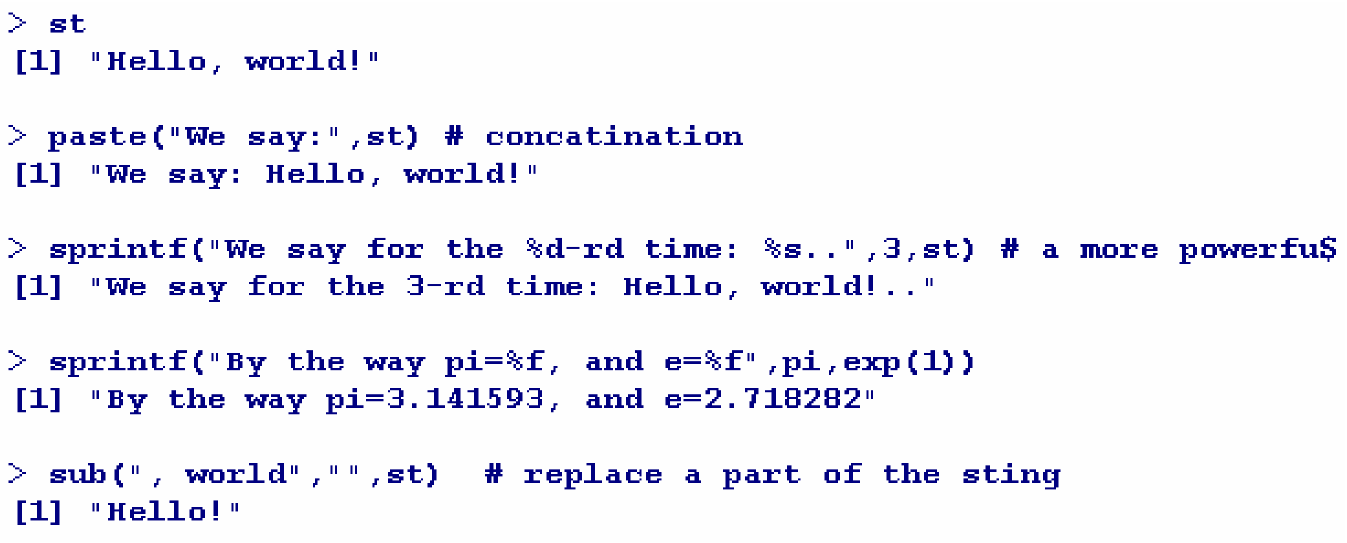


```
> l=pi*2*r # let us calculate the

> l
[1] 9.424778

> b1=TRUE    # try  b1=T

> b2=FALSE   # try  b2=F

> b1 & b2    # logical AND
[1] FALSE

> b1 | b2    # logical OR
[1] TRUE

> !b1        # logical AND
[1] FALSE

> xor(b1,b2)# logical XOR
[1] TRUE

> r==l
[1] FALSE

> r<l
[1] TRUE
> |
```

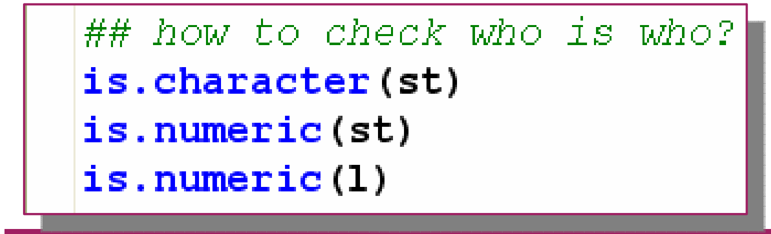
4. BASIC OPERATIONS (4.2)



```
##---------------------------
## Character (strings)
st = "Hello, world!"
st
paste("We say:",st) # concatination
sprintf("We say for the %d-rd time: %s..",3,st) # a more powerfull method a-la C
sprintf("By the way pi=%f, and e=%f",pi,exp(1))

sub(", world","",st)   # replace a part of the sting
                       # (*) in R the regular expression are used to define the patte
casefold(st, upper=T)  # change the case
nchar(st)              # number of characters
strsplit(st,"")[[1]]   # (*) transforms a string into the vector of single characters
```

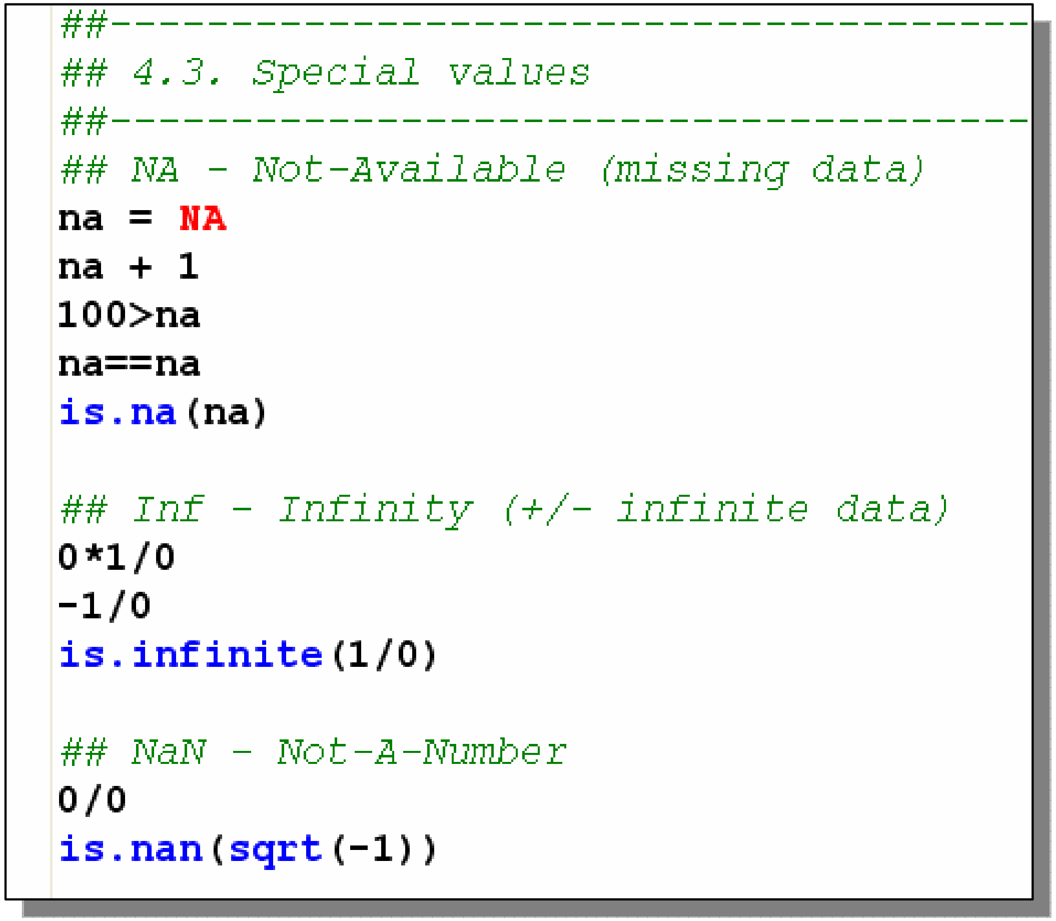


```
> st
[1] "Hello, world!"

> paste("We say:",st) # concatination
[1] "We say: Hello, world!"

> sprintf("We say for the %d-rd time: %s..",3,st) # a more powerfu$
[1] "We say for the 3-rd time: Hello, world!.."

> sprintf("By the way pi=%f, and e=%f",pi,exp(1))
[1] "By the way pi=3.141593, and e=2.718282"

> sub(", world","",st)  # replace a part of the sting
[1] "Hello!"
```

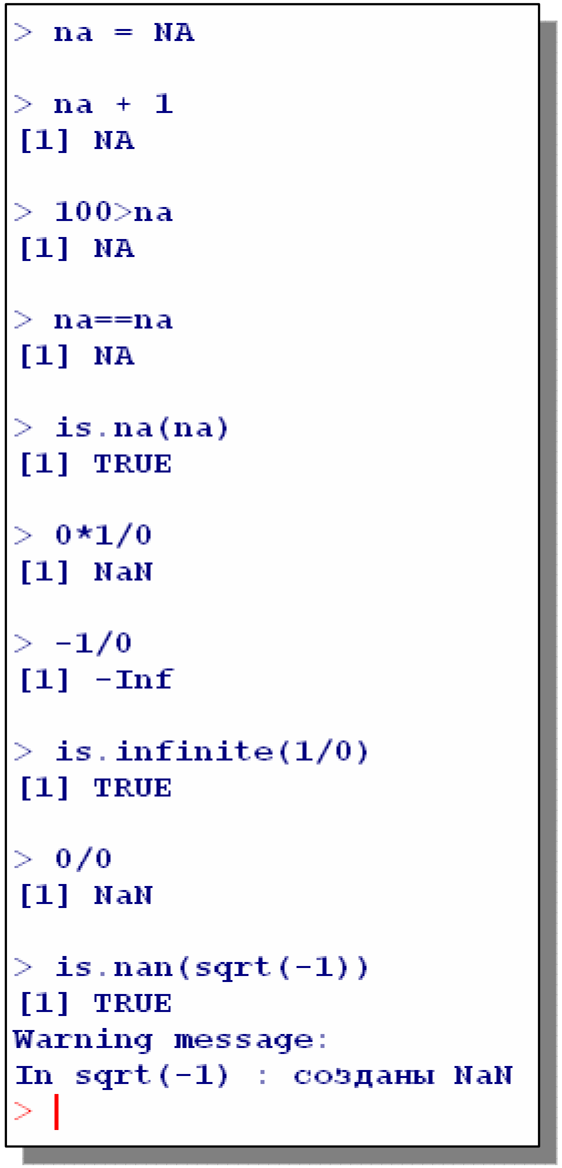


```
## how to check who is who?
is.character(st)
is.numeric(st)
is.numeric(1)
```

4. BASIC OPERATIONS (4.3)



```
##-----------------------------------------
## 4.3. Special values
##-----------------------------------------
## NA - Not-Available (missing data)
na = NA
na + 1
100>na
na==na
is.na(na)

## Inf - Infinity (+/- infinite data)
0*1/0
-1/0
is.infinite(1/0)

## NaN - Not-A-Number
0/0
is.nan(sqrt(-1))
```

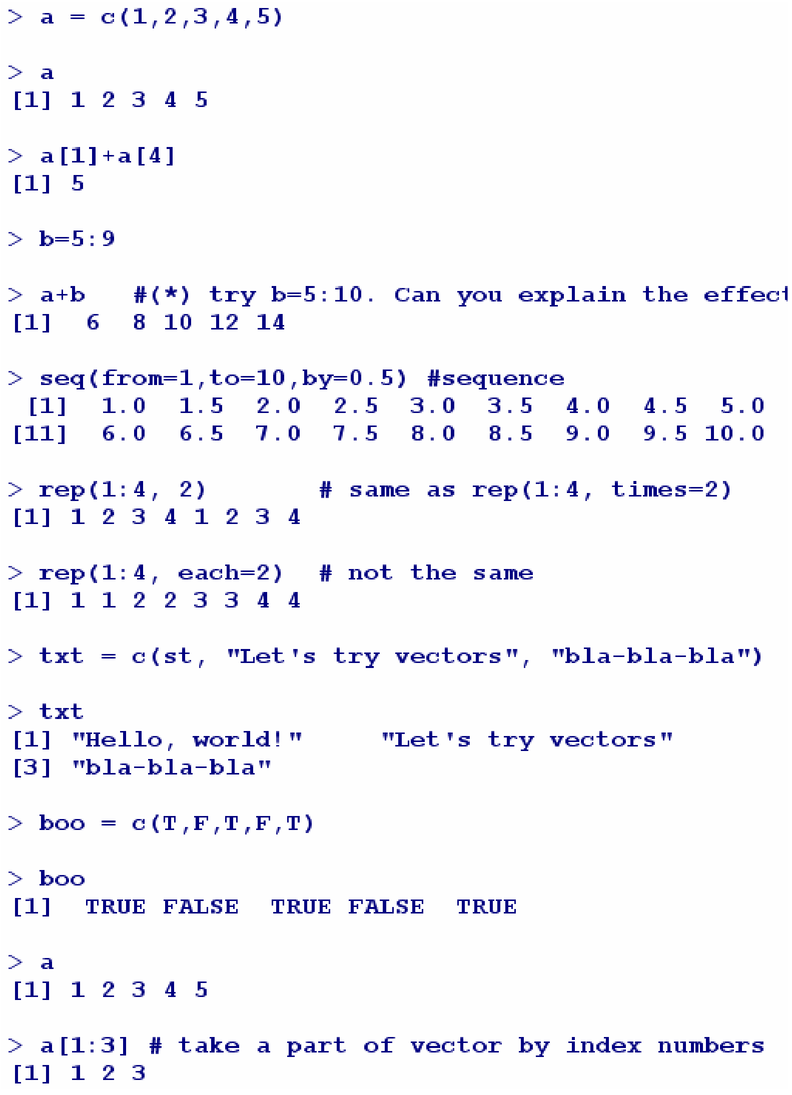


```
> na = NA

> na + 1
[1] NA

> 100>na
[1] NA

> na==na
[1] NA

> is.na(na)
[1] TRUE

> 0*1/0
[1] NaN

> -1/0
[1] -Inf

> is.infinite(1/0)
[1] TRUE

> 0/0
[1] NaN

> is.nan(sqrt(-1))
[1] TRUE
Warning message:
In sqrt(-1) : созданы NaN
> |
```

4. BASIC OPERATIONS (4.4)

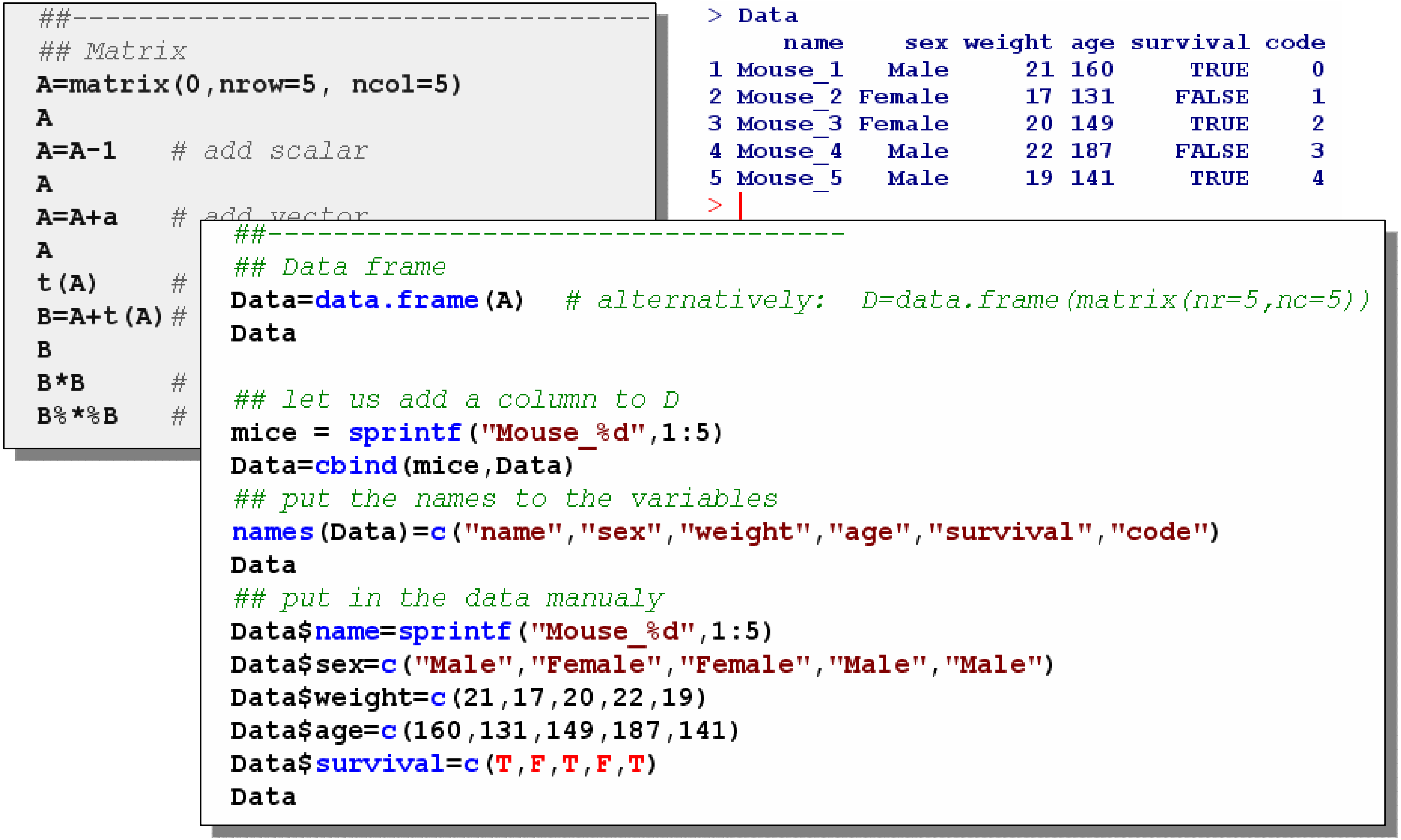
```
##-----------------------------------
## 4.4. Vectors, matrixes and lists
##-----------------------------------
```

```
## Vector creation
a = c(1,2,3,4,5)
a
a[1]+a[4]

b=5:9
a+b   #(*) try b=5:10. Can you explain the effect?

seq(from=1,to=10,by=0.5) #sequence
rep(1:4, 2)         # same as rep(1:4, times=2)
rep(1:4, each=2)   # not the same

txt = c(st, "Let's try vectors", "bla-bla-bla")
txt

boo = c(T,F,T,F,T)
boo

## Vector indexes
a
a[1:3] # take a part of vector by index numbers
a[boo] # take a part of vector by logical vector
a[a>2] # take a part by a condition (!!!)
```



```
> a = c(1,2,3,4,5)

> a
[1] 1 2 3 4 5

> a[1]+a[4]
[1] 5

> b=5:9

> a+b   #(*) try b=5:10. Can you explain the effec
[1]  6  8 10 12 14

> seq(from=1,to=10,by=0.5) #sequence
 [1]  1.0  1.5  2.0  2.5  3.0  3.5  4.0  4.5  5.0
[11]  6.0  6.5  7.0  7.5  8.0  8.5  9.0  9.5 10.0

> rep(1:4, 2)       # same as rep(1:4, times=2)
[1] 1 2 3 4 1 2 3 4

> rep(1:4, each=2)  # not the same
[1] 1 1 2 2 3 3 4 4

> txt = c(st, "Let's try vectors", "bla-bla-bla")

> txt
[1] "Hello, world!"     "Let's try vectors"
[3] "bla-bla-bla"

> boo = c(T,F,T,F,T)

> boo
[1]  TRUE FALSE  TRUE FALSE  TRUE

> a
[1] 1 2 3 4 5

> a[1:3] # take a part of vector by index numbers
[1] 1 2 3
```

4. BASIC OPERATIONS (4.4)



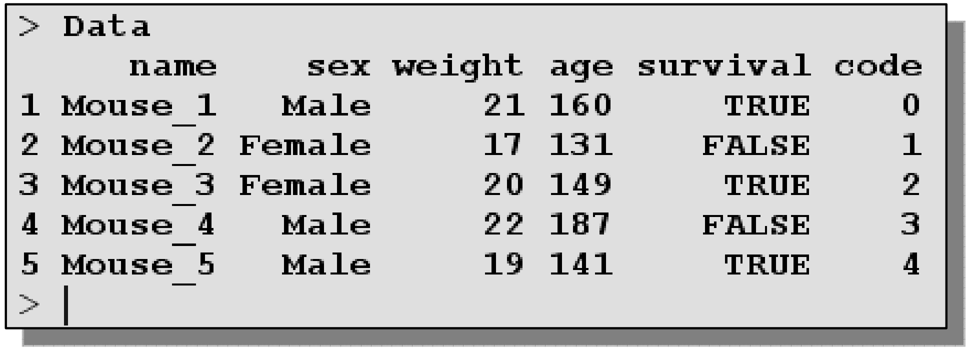
```
##------------------------------------
## Matrix
A=matrix(0,nrow=5, ncol=5)
A
A=A-1    # add scalar
A
A=A+a    # add vector
A
t(A)     #
B=A+t(A) #
B
B*B      #
B%*%B    #
```

```
> Data
     name    sex weight age survival code
1 Mouse_1   Male     21 160     TRUE    0
2 Mouse_2 Female     17 131    FALSE    1
3 Mouse_3 Female     20 149     TRUE    2
4 Mouse_4   Male     22 187    FALSE    3
5 Mouse_5   Male     19 141     TRUE    4
>
```

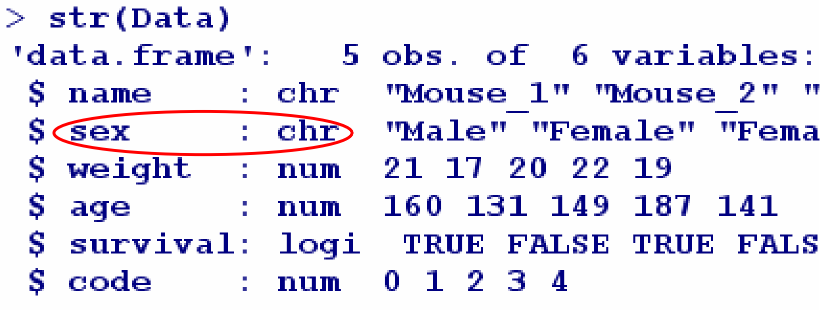
```
##-----------------------------------
## Data frame
Data=data.frame(A)  # alternatively:  D=data.frame(matrix(nr=5,nc=5))
Data

## let us add a column to D
mice = sprintf("Mouse_%d",1:5)
Data=cbind(mice,Data)
## put the names to the variables
names(Data)=c("name","sex","weight","age","survival","code")
Data
## put in the data manualy
Data$name=sprintf("Mouse_%d",1:5)
Data$sex=c("Male","Female","Female","Male","Male")
Data$weight=c(21,17,20,22,19)
Data$age=c(160,131,149,187,141)
Data$survival=c(T,F,T,F,T)
Data
```

4. BASIC OPERATIONS (4.4)

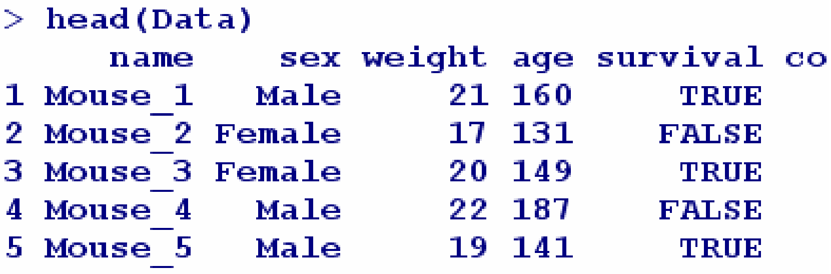
```
## (!!!) see the structure of the objects
str(Data)

## see the structure of the objects
head(Data)

## summary on the data
summary(Data)

## Let's use factors
Data$sex = factor(Data$sex)
summary(Data)

## usefull commands when working with factors:
levels(Data$sex)        # returns levels of the factor
nlevels(Data$sex)       # returns number of levels
as.character(Data$sex)  # transform into strings
```



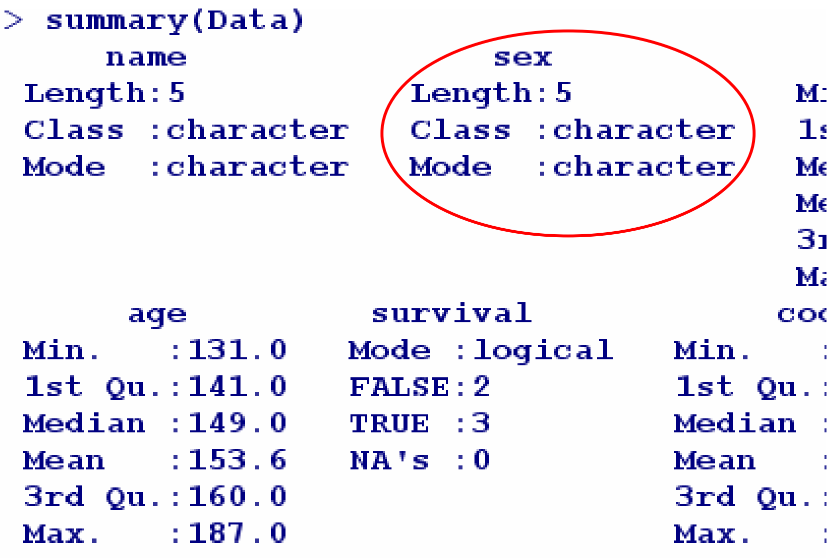
```
> Data
     name    sex weight age survival code
1 Mouse_1   Male     21 160     TRUE    0
2 Mouse_2 Female     17 131    FALSE    1
3 Mouse_3 Female     20 149     TRUE    2
4 Mouse_4   Male     22 187    FALSE    3
5 Mouse_5   Male     19 141     TRUE    4
> |
```



```
> str(Data)
'data.frame':   5 obs. of  6 variables:
 $ name    : chr  "Mouse_1" "Mouse_2" "
 $ sex     : chr  "Male" "Female" "Fema
 $ weight  : num  21 17 20 22 19
 $ age     : num  160 131 149 187 141
 $ survival: logi  TRUE FALSE TRUE FALS
 $ code    : num  0 1 2 3 4
```

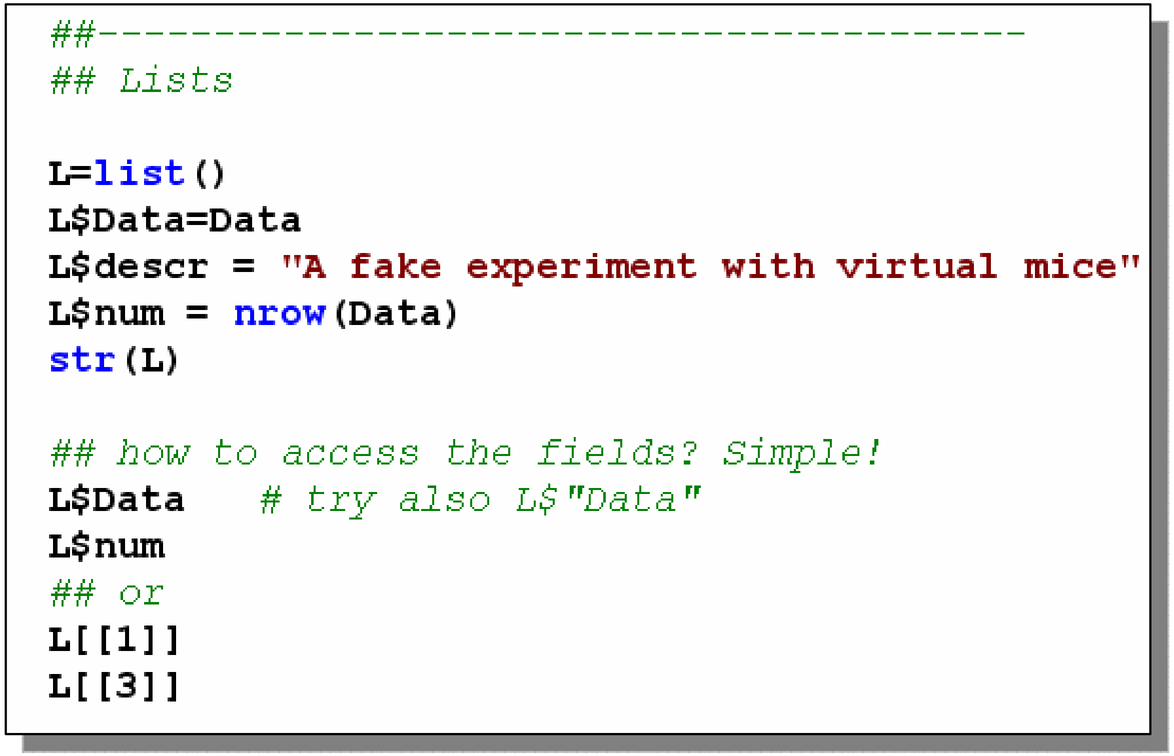


```
> head(Data)
     name    sex weight age survival co
1 Mouse_1   Male     21 160     TRUE
2 Mouse_2 Female     17 131    FALSE
3 Mouse_3 Female     20 149     TRUE
4 Mouse_4   Male     22 187    FALSE
5 Mouse_5   Male     19 141     TRUE
```

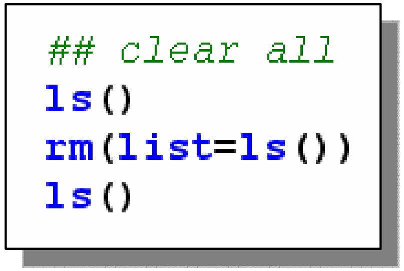


```
> summary(Data)
     name               sex                M:
 Length:5           Length:5           1s
 Class :character   Class :character   Me
 Mode  :character   Mode  :character   Me
                                       3r
                                       Ma
      age          survival          coo
 Min.   :131.0   Mode :logical   Min.   :
 1st Qu.:141.0   FALSE:2         1st Qu.:
 Median :149.0   TRUE :3         Median :
 Mean   :153.6   NA's :0         Mean   :
 3rd Qu.:160.0                   3rd Qu.:
 Max.   :187.0                   Max.   :
```

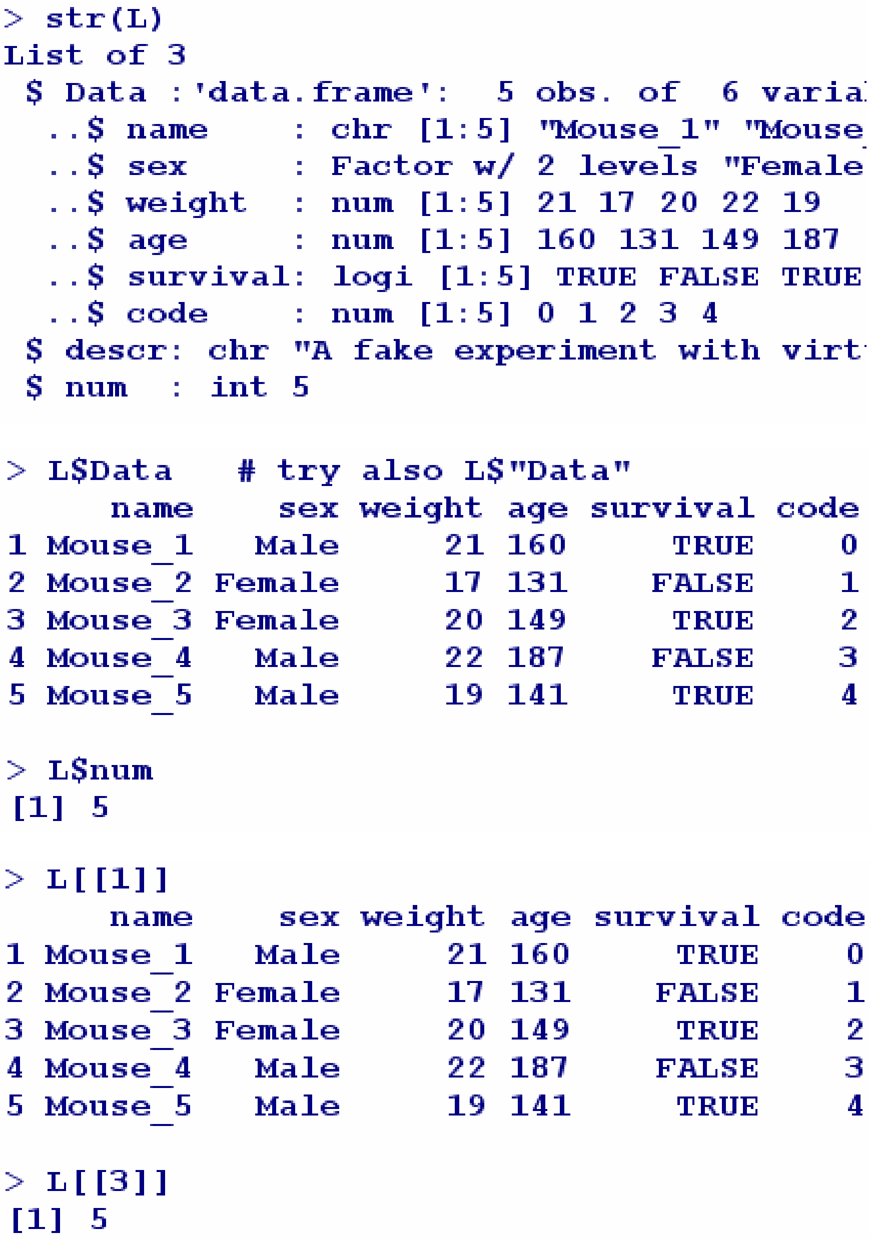
4. BASIC OPERATIONS (4.4)



```
##-----------------------------------------
## Lists

L=list()
L$Data=Data
L$descr = "A fake experiment with virtual mice"
L$num = nrow(Data)
str(L)

## how to access the fields? Simple!
L$Data    # try also L$"Data"
L$num
## or
L[[1]]
L[[3]]
```



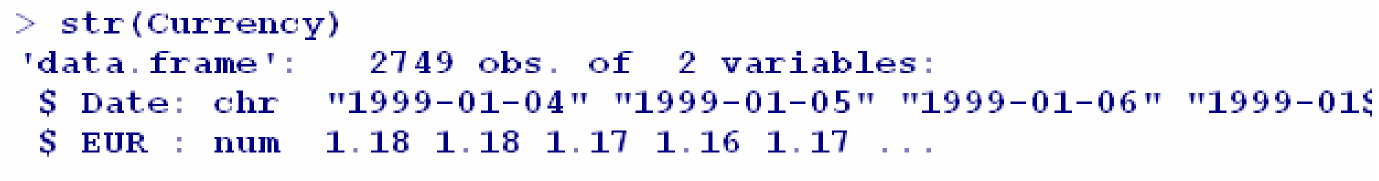
```
## clear all
ls()
rm(list=ls())
ls()
```



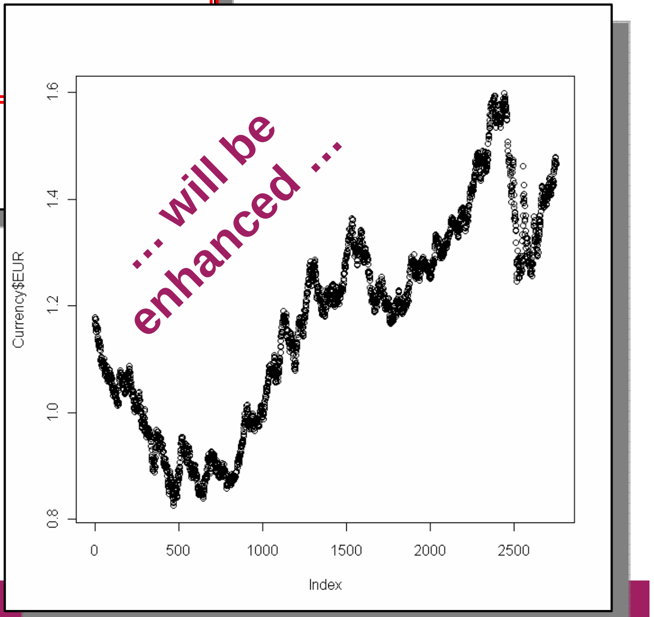
```
> str(L)
List of 3
 $ Data :'data.frame':  5 obs. of  6 varia
  ..$ name    : chr [1:5] "Mouse_1" "Mouse
  ..$ sex     : Factor w/ 2 levels "Female
  ..$ weight  : num [1:5] 21 17 20 22 19
  ..$ age     : num [1:5] 160 131 149 187
  ..$ survival: logi [1:5] TRUE FALSE TRUE
  ..$ code    : num [1:5] 0 1 2 3 4
 $ descr: chr "A fake experiment with virt
 $ num  : int 5

> L$Data    # try also L$"Data"
     name    sex weight age survival code
1 Mouse_1   Male     21 160     TRUE    0
2 Mouse_2 Female     17 131    FALSE    1
3 Mouse_3 Female     20 149     TRUE    2
4 Mouse_4   Male     22 187    FALSE    3
5 Mouse_5   Male     19 141     TRUE    4

> L$num
[1] 5

> L[[1]]
     name    sex weight age survival code
1 Mouse_1   Male     21 160     TRUE    0
2 Mouse_2 Female     17 131    FALSE    1
3 Mouse_3 Female     20 149     TRUE    2
4 Mouse_4   Male     22 187    FALSE    3
5 Mouse_5   Male     19 141     TRUE    4

> L[[3]]
[1] 5
```

5. DATA IMPORT AND EXPORT (5.1)

```
##--------------------------------------------------------------
## "Currency" dataset: learn how to use "header" and "as.is"
##--------------------------------------------------------------
Currency = read.table("http://edu.sablab.net/r/data/currency.txt")
str(Currency)
head(Currency)
## hmm.. wrong... let's ask to add header
Currency = read.table("http://edu.sablab.net/r/data/currency.txt",
                      header=T)
str(Currency)
head(Currency)
## still, it's better to consider dates as strings, not as factors
Currency = read.table("http://edu.sablab.net/r/data/currency.txt",
                      header=T,as.is=T)
str(Currency)
summary(Currency)
## correct!
plot(Currency$EUR)
```



```
> str(Currency)
'data.frame':   2749 obs. of  2 variables:
 $ Date: chr  "1999-01-04" "1999-01-05" "1999-01-06" "1999-01$
 $ EUR : num  1.18 1.18 1.17 1.16 1.17 ...
```



... will be enhanced ...

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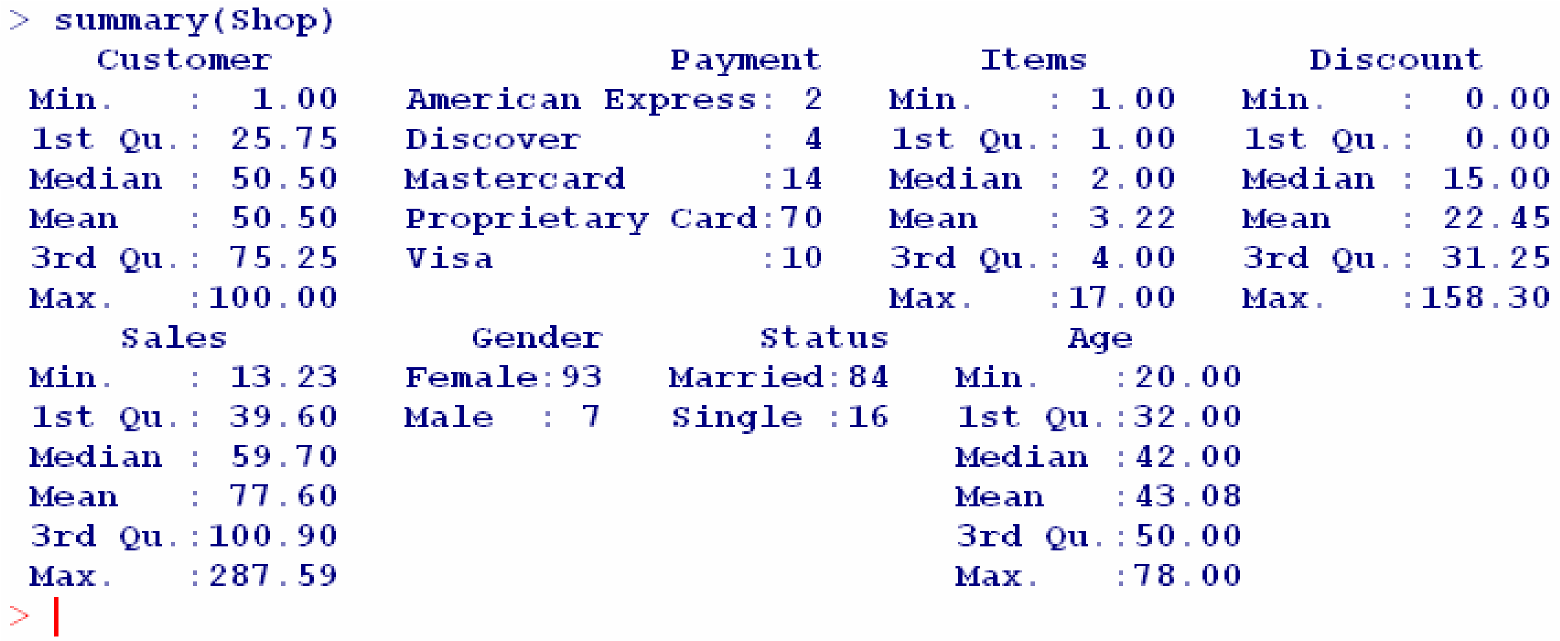
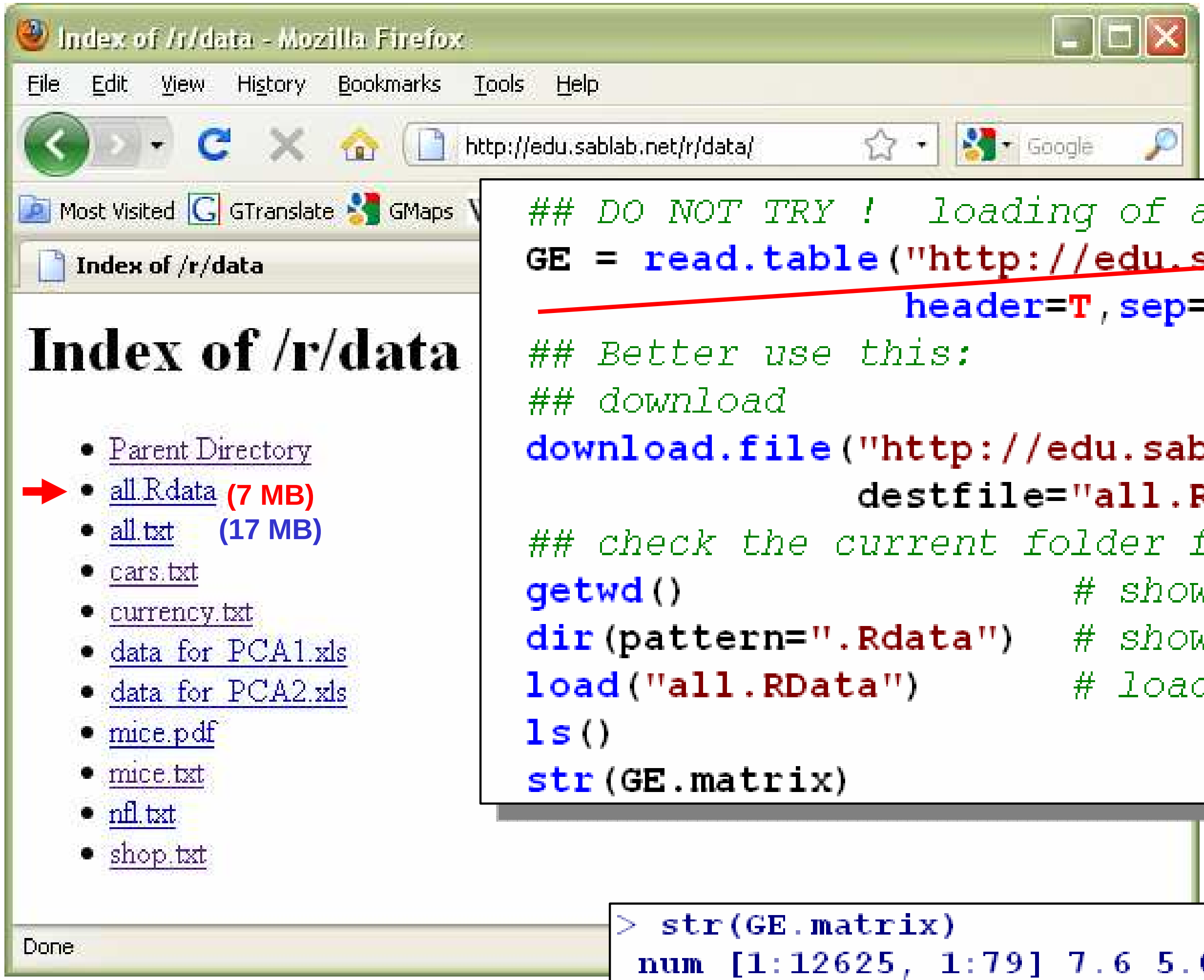
5. DATA IMPORT AND EXPORT (5.2)

```
##----------------------------------------------------------------
## 5.2. "Shop" dataset: learn how to use "sep"
##----------------------------------------------------------------
Shop = read.table("http://edu.sablab.net/r/data/shop.txt",
                  header=T)
## hm... an error!
Shop = read.table("http://edu.sablab.net/r/data/shop.txt",
                  header=T,sep="\t")
## correct!
str(Shop)
head(Shop)
```



```
> summary(Shop)
    Customer                      Payment         Items           Discount     
 Min.   :  1.00   American Express: 2   Min.   : 1.00   Min.   :  0.00  
 1st Qu.: 25.75   Discover        : 4   1st Qu.: 1.00   1st Qu.:  0.00  
 Median : 50.50   Mastercard      :14   Median : 2.00   Median : 15.00  
 Mean   : 50.50   Proprietary Card:70   Mean   : 3.22   Mean   : 22.45  
 3rd Qu.: 75.25   Visa            :10   3rd Qu.: 4.00   3rd Qu.: 31.25  
 Max.   :100.00                         Max.   :17.00   Max.   :158.30  
     Sales            Gender       Status         Age       
 Min.   : 13.23   Female:93   Married:84   Min.   :20.00  
 1st Qu.: 39.60   Male  : 7   Single :16   1st Qu.:32.00  
 Median : 59.70                            Median :42.00  
 Mean   : 77.60                            Mean   :43.08  
 3rd Qu.:100.90                            3rd Qu.:50.00  
 Max.   :287.59                            Max.   :78.00  
> 
```

5. DATA IMPORT AND EXPORT (5.3)


Index of /r/data - Mozilla Firefox
File Edit View History Bookmarks Tools Help
http://edu.sablab.net/r/data/
Google
Most Visited
GTranslate
GMaps
Index of /r/data
Done

Index of /r/data

- Parent Directory
- all.Rdata **(7 MB)**
- all.txt **(17 MB)**
- cars.txt
- currency.txt
- data_for_PCA1.xls
- data_for_PCA2.xls
- mice.pdf
- mice.txt
- nfl.txt
- shop.txt

```
## DO NOT TRY !  loading of a big matrix
~~GE = read.table("http://edu.sablab.net/r/data/all.txt",
               header=T,sep="\t")~~
## Better use this:
## download
download.file("http://edu.sablab.net/r/data/all.Rdata",
              destfile="all.Rdata",mode = "wb")
## check the current folder for ".Rdata" file
getwd()                # show current folder
dir(pattern=".Rdata")  # show files in the current folder
load("all.RData")      # load the data
ls()
str(GE.matrix)
```

```
> str(GE.matrix)
 num [1:12625, 1:79] 7.6 5.05 3.9 5.9 5.93 ...
 - attr(*, "dimnames")=List of 2
  ..$ : chr [1:12625] "MAPK3" "TIE1" "CYP2C19" "CXCR5"
  ..$ : chr [1:79] "BCRABL_01" "BCRABL_03" "BCRABL_09"
> |
```

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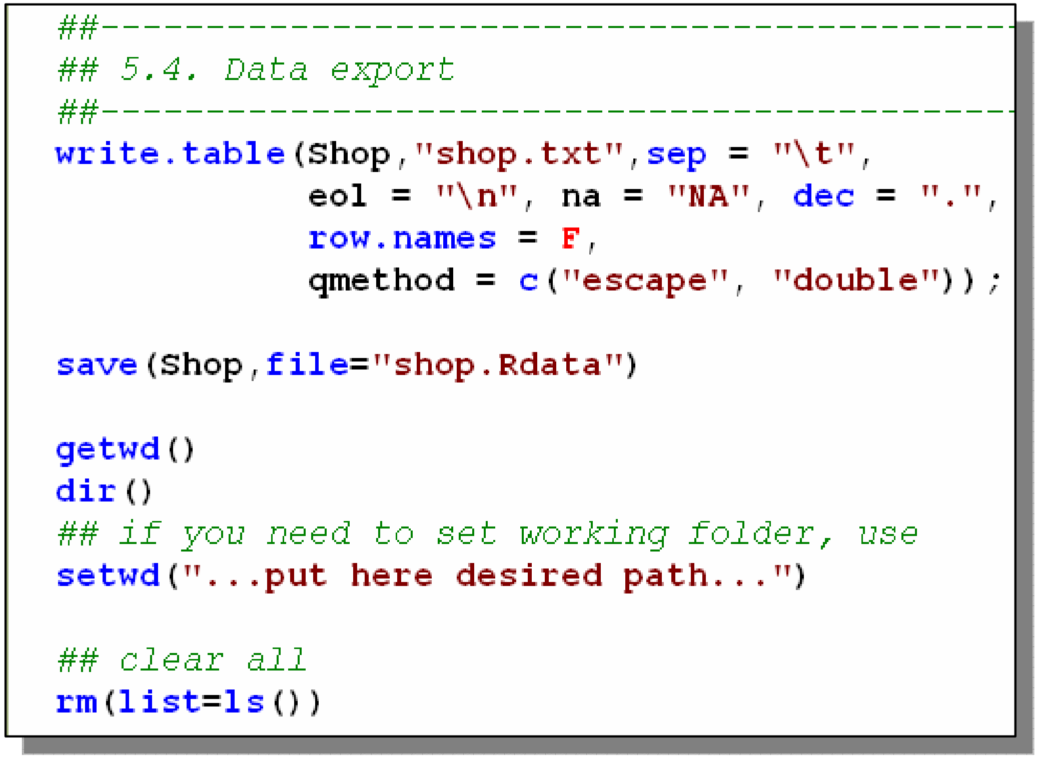
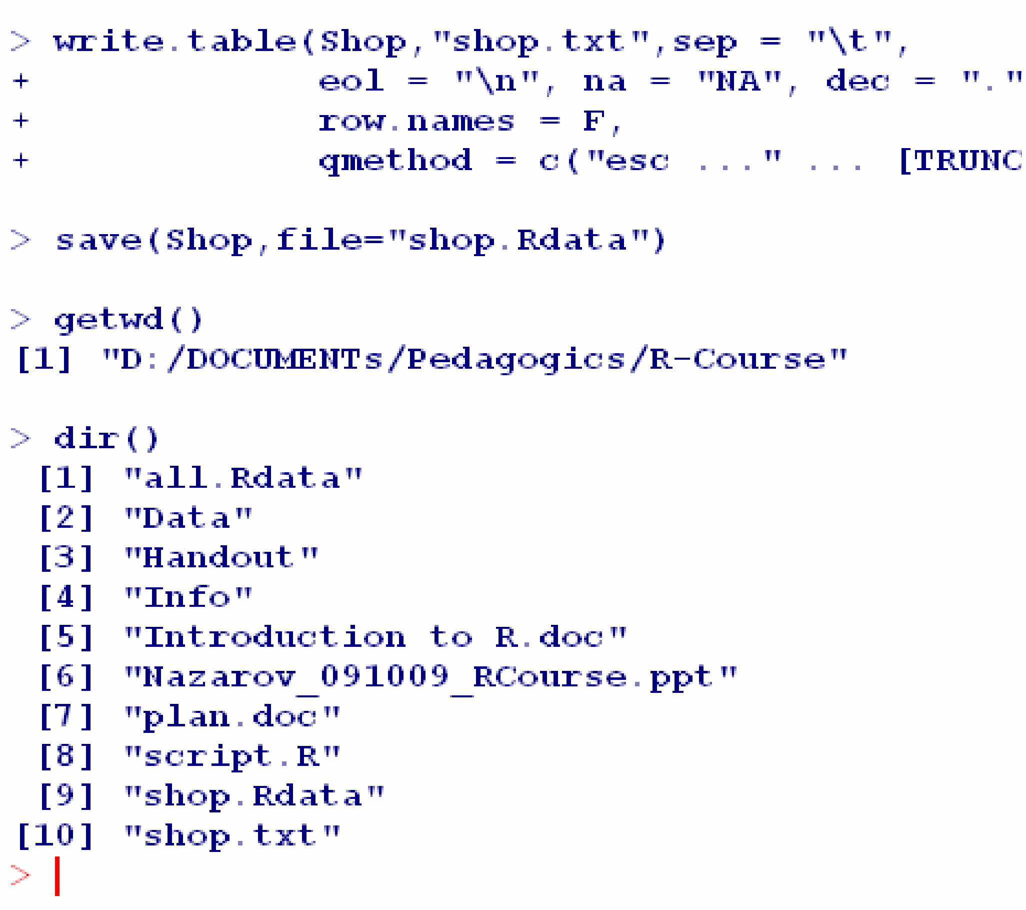
5. DATA IMPORT AND EXPORT (5.4)

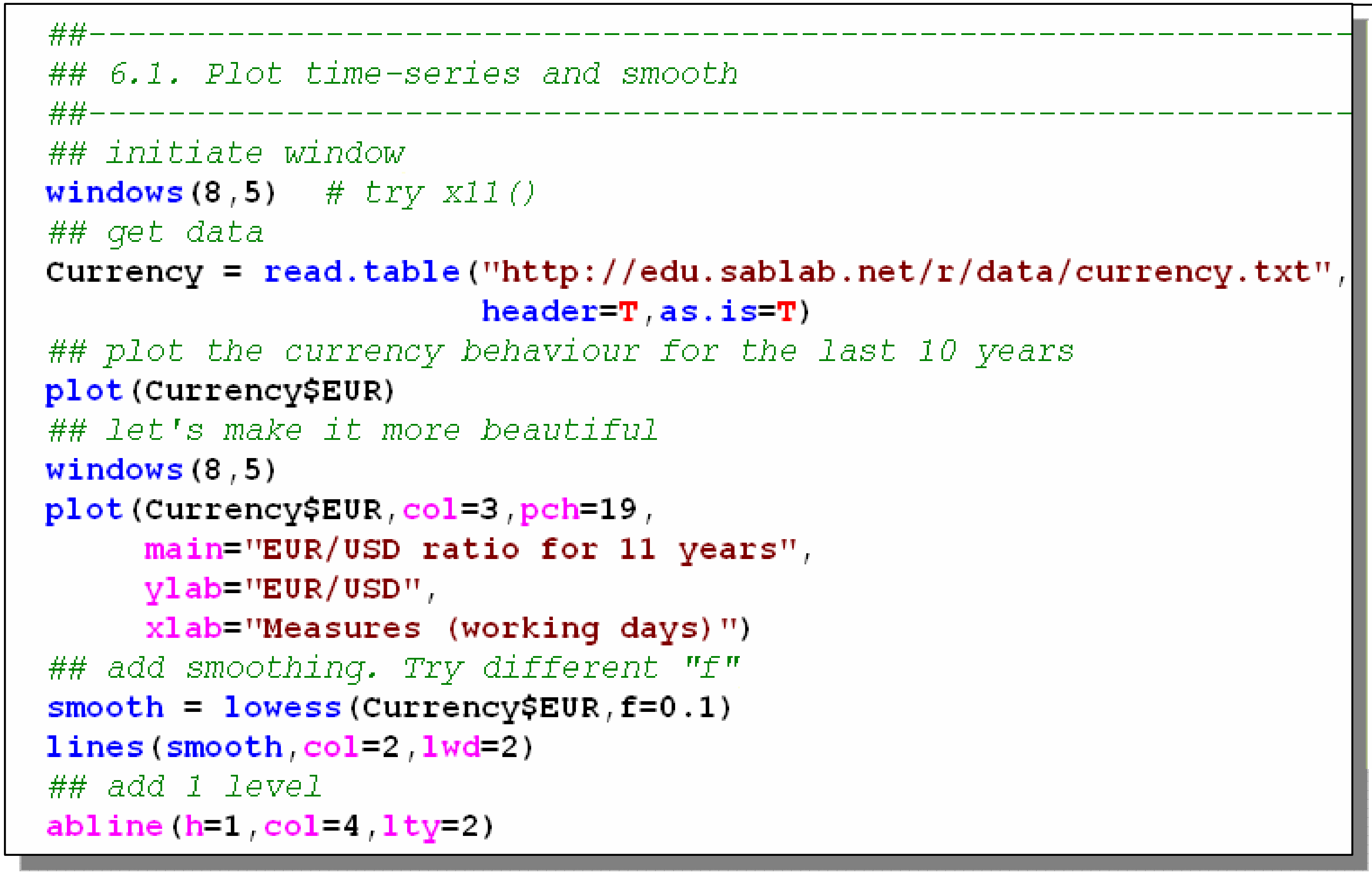


```
##-------------------------------------------------------
## 5.4. Data export
##-------------------------------------------------------
write.table(Shop,"shop.txt",sep = "\t",
            eol = "\n", na = "NA", dec = ".",
            row.names = F,
            qmethod = c("escape", "double"));

save(Shop,file="shop.Rdata")

getwd()
dir()
## if you need to set working folder, use
setwd("...put here desired path...")

## clear all
rm(list=ls())
```

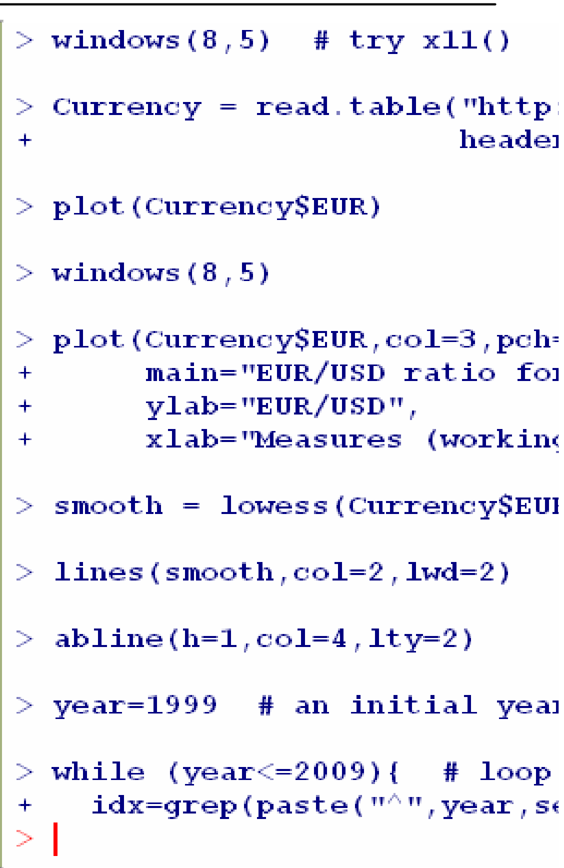


```
> write.table(Shop,"shop.txt",sep = "\t",
+             eol = "\n", na = "NA", dec = "."
+             row.names = F,
+             qmethod = c("esc ..." ... [TRUNC

> save(Shop,file="shop.Rdata")

> getwd()
[1] "D:/DOCUMENTs/Pedagogics/R-Course"

> dir()
 [1] "all.Rdata"
 [2] "Data"
 [3] "Handout"
 [4] "Info"
 [5] "Introduction to R.doc"
 [6] "Nazarov_091009_RCourse.ppt"
 [7] "plan.doc"
 [8] "script.R"
 [9] "shop.Rdata"
[10] "shop.txt"
> 
```

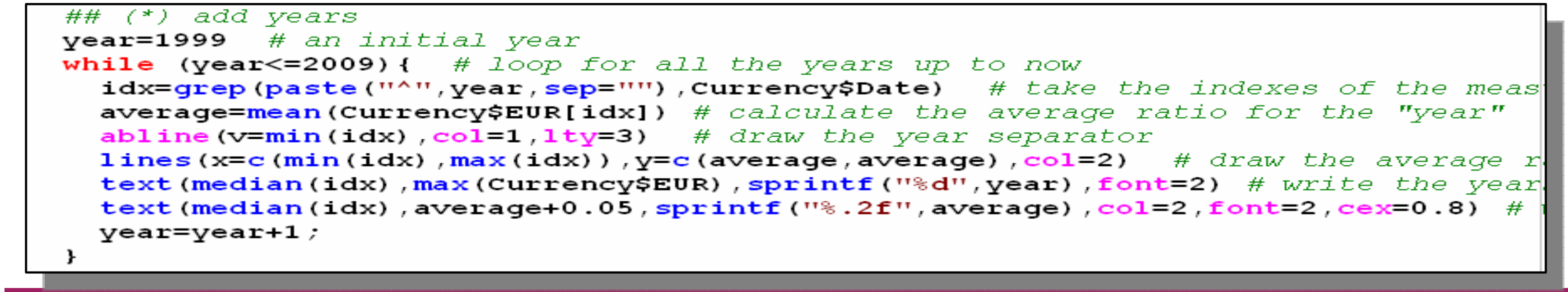
6. DATA VISUALIZATION (6.1)



```
##--------------------------------------------------------------------
## 6.1. Plot time-series and smooth
##--------------------------------------------------------------------
## initiate window
windows(8,5)   # try x11()
## get data
Currency = read.table("http://edu.sablab.net/r/data/currency.txt",
                      header=T,as.is=T)
## plot the currency behaviour for the last 10 years
plot(Currency$EUR)
## let's make it more beautiful
windows(8,5)
plot(Currency$EUR,col=3,pch=19,
     main="EUR/USD ratio for 11 years",
     ylab="EUR/USD",
     xlab="Measures (working days)")
## add smoothing. Try different "f"
smooth = lowess(Currency$EUR,f=0.1)
lines(smooth,col=2,lwd=2)
## add 1 level
abline(h=1,col=4,lty=2)
```

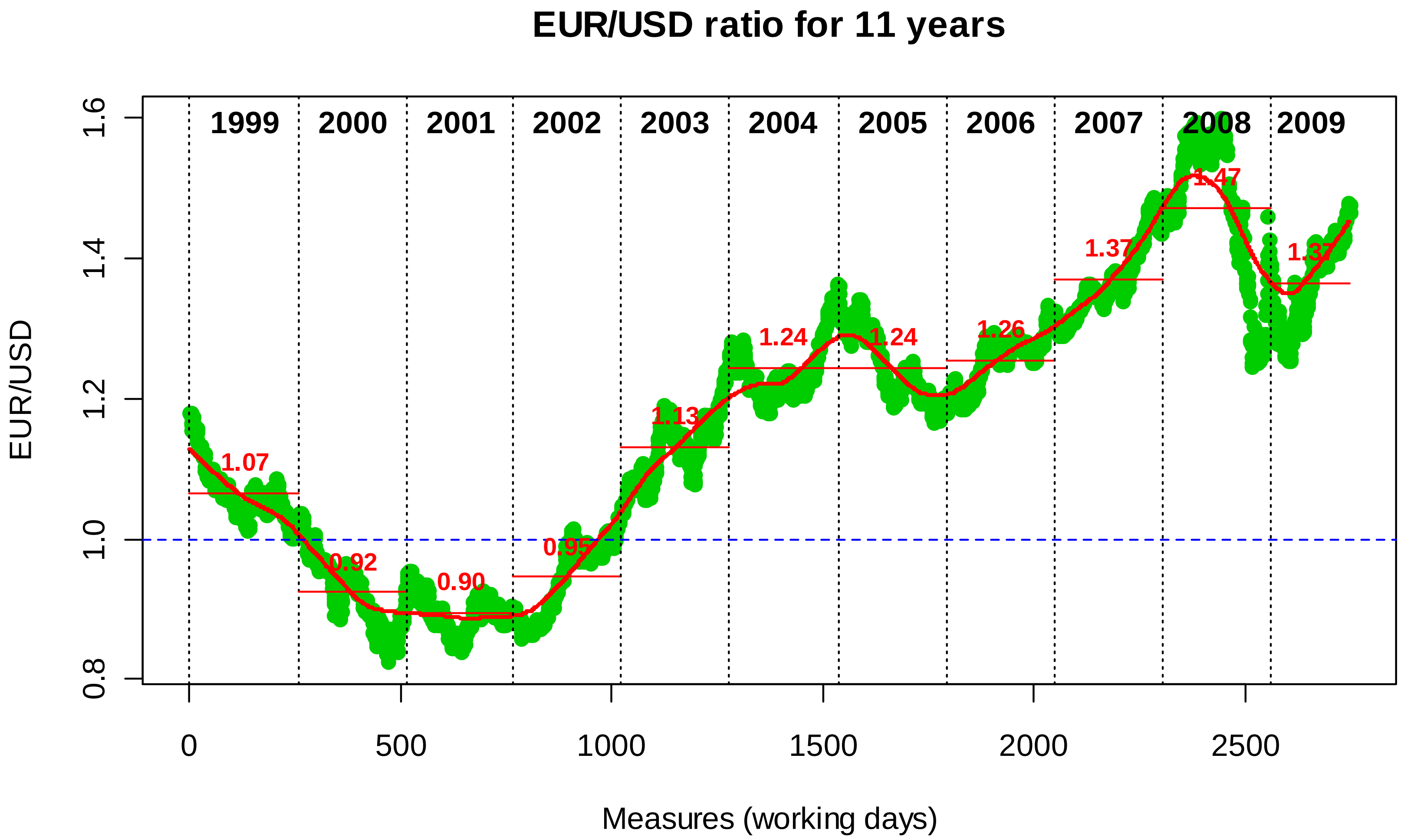


```
> windows(8,5)   # try x11()

> Currency = read.table("http:
+                           header

> plot(Currency$EUR)

> windows(8,5)

> plot(Currency$EUR,col=3,pch=
+       main="EUR/USD ratio for
+       ylab="EUR/USD",
+       xlab="Measures (working

> smooth = lowess(Currency$EUR

> lines(smooth,col=2,lwd=2)

> abline(h=1,col=4,lty=2)

> year=1999  # an initial year

> while (year<=2009){   # loop
+   idx=grep(paste("^",year,se
> |
```



```
## (*) add years
year=1999  # an initial year
while (year<=2009){  # loop for all the years up to now
  idx=grep(paste("^",year,sep=""),Currency$Date)  # take the indexes of the meas
  average=mean(Currency$EUR[idx]) # calculate the average ratio for the "year"
  abline(v=min(idx),col=1,lty=3)  # draw the year separator
  lines(x=c(min(idx),max(idx)),y=c(average,average),col=2)  # draw the average r
  text(median(idx),max(Currency$EUR),sprintf("%d",year),font=2) # write the year
  text(median(idx),average+0.05,sprintf("%.2f",average),col=2,font=2,cex=0.8) # 
  year=year+1;
}
```

6. DATA VISUALIZATION (6.1)



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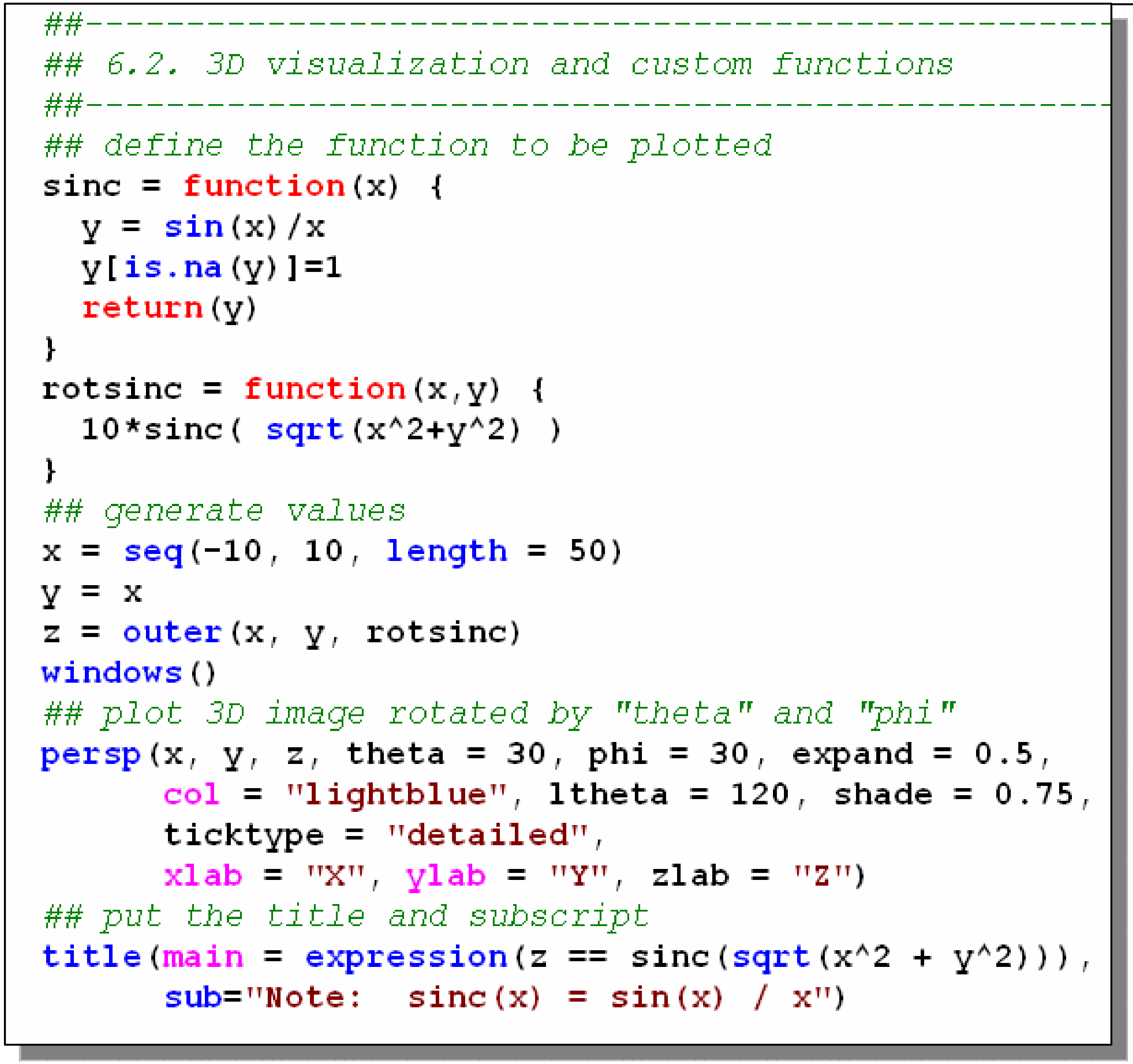
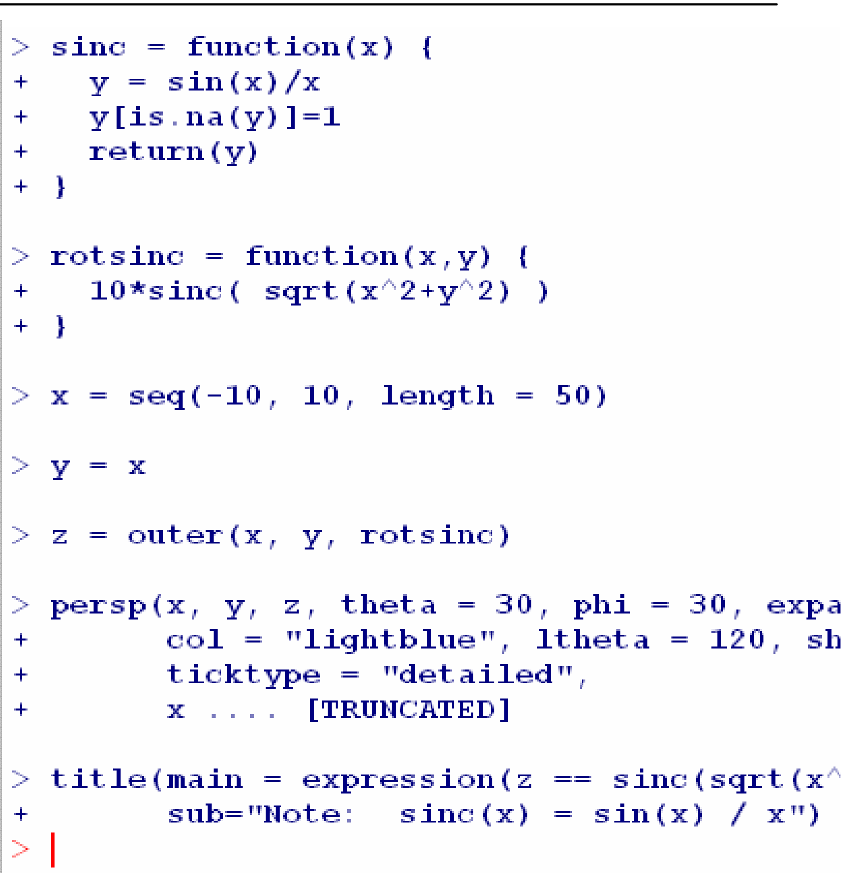
6. DATA VISUALIZATION (6.2)



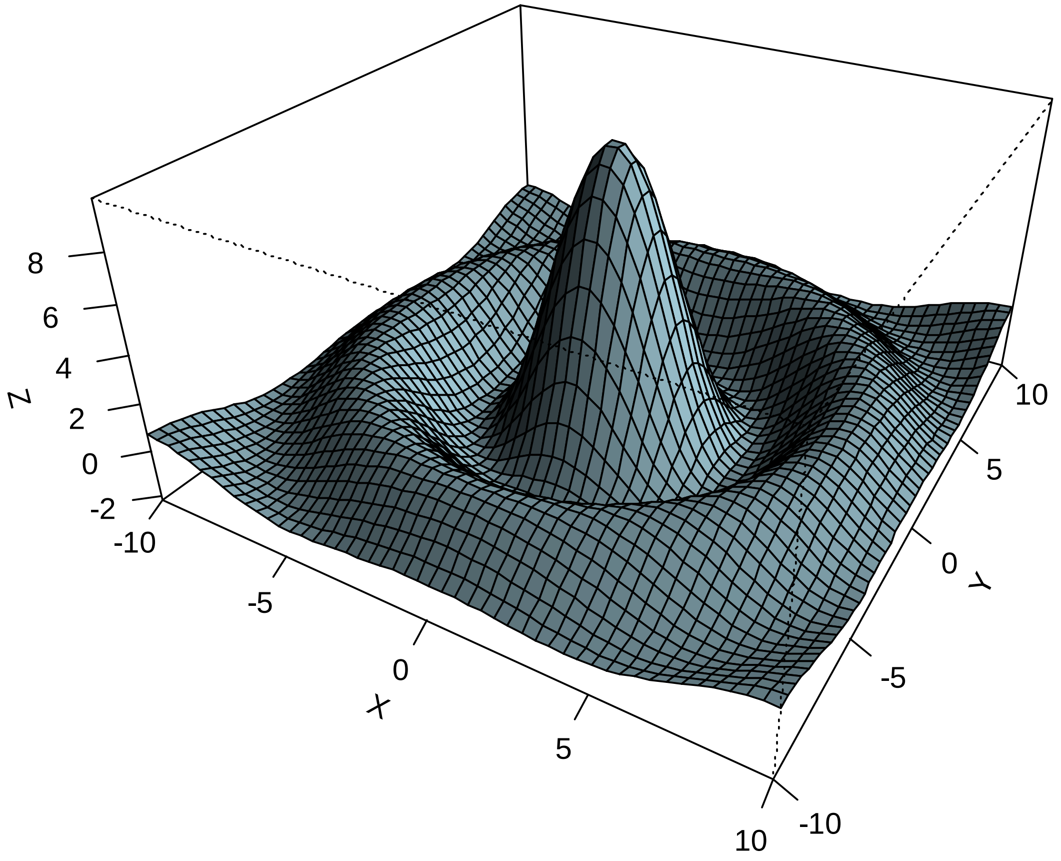
```
##-----------------------------------------------------
## 6.2. 3D visualization and custom functions
##-----------------------------------------------------
## define the function to be plotted
sinc = function(x) {
  y = sin(x)/x
  y[is.na(y)]=1
  return(y)
}
rotsinc = function(x,y) {
  10*sinc( sqrt(x^2+y^2) )
}
## generate values
x = seq(-10, 10, length = 50)
y = x
z = outer(x, y, rotsinc)
windows()
## plot 3D image rotated by "theta" and "phi"
persp(x, y, z, theta = 30, phi = 30, expand = 0.5,
      col = "lightblue", ltheta = 120, shade = 0.75,
      ticktype = "detailed",
      xlab = "X", ylab = "Y", zlab = "Z")
## put the title and subscript
title(main = expression(z == sinc(sqrt(x^2 + y^2))),
      sub="Note:  sinc(x) = sin(x) / x")
```



```
> sinc = function(x) {
+   y = sin(x)/x
+   y[is.na(y)]=1
+   return(y)
+ }

> rotsinc = function(x,y) {
+   10*sinc( sqrt(x^2+y^2) )
+ }

> x = seq(-10, 10, length = 50)

> y = x

> z = outer(x, y, rotsinc)

> persp(x, y, z, theta = 30, phi = 30, expa
+       col = "lightblue", ltheta = 120, sh
+       ticktype = "detailed",
+       x .... [TRUNCATED]

> title(main = expression(z == sinc(sqrt(x^
+       sub="Note:  sinc(x) = sin(x) / x")
> 
```

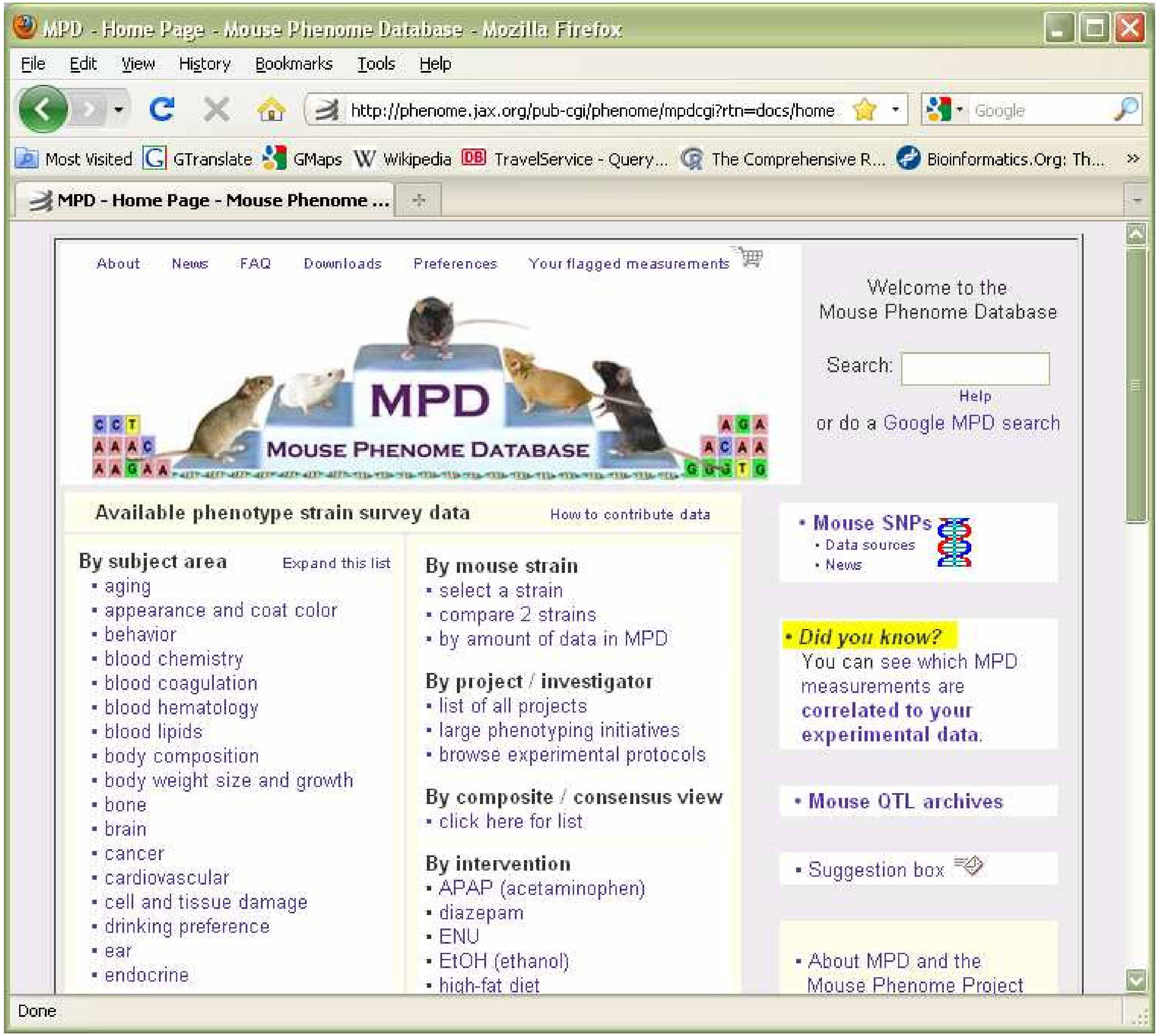
6. DATA VISUALIZATION (6.2)

$$z = \mathrm{sinc}(\sqrt{x^2 + y^2})$$



Note: sinc(x) = sin(x) / x

6. DATA VISUALIZATION (6.3)


MPD - Home Page - Mouse Phenome Database - Mozilla Firefox
File Edit View History Bookmarks Tools Help
http://phenome.jax.org/pub-cgi/phenome/mpdcgi?rtn=docs/home
Google
Most Visited GTranslate GMaps Wikipedia TravelService - Query... The Comprehensive R... Bioinformatics.Org: Th...
MPD - Home Page - Mouse Phenome ...
About News FAQ Downloads Preferences Your flagged measurements
MPD
MOUSE PHENOME DATABASE
Done

Welcome to the Mouse Phenome Database

Search:

Help

or do a Google MPD search

Available phenotype strain survey data

How to contribute data

By subject area — Expand this list
- aging
- appearance and coat color
- behavior
- blood chemistry
- blood coagulation
- blood hematology
- blood lipids
- body composition
- body weight size and growth
- bone
- brain
- cancer
- cardiovascular
- cell and tissue damage
- drinking preference
- ear
- endocrine

By mouse strain
- select a strain
- compare 2 strains
- by amount of data in MPD

By project / investigator
- list of all projects
- large phenotyping initiatives
- browse experimental protocols

By composite / consensus view
- click here for list

By intervention
- APAP (acetaminophen)
- diazepam
- ENU
- EtOH (ethanol)
- high-fat diet

- Mouse SNPs
  - Data sources
  - News

- *Did you know?* You can see which MPD measurements are **correlated to your experimental data.**

- Mouse QTL archives

- Suggestion box

- About MPD and the Mouse Phenome Project

Tordoff MG, Bachmanov AA

Survey of calcium & sodium intake and metabolism with bone and body composition data

Project symbol: **Tordoff3**

Accession number: **MPD:103**

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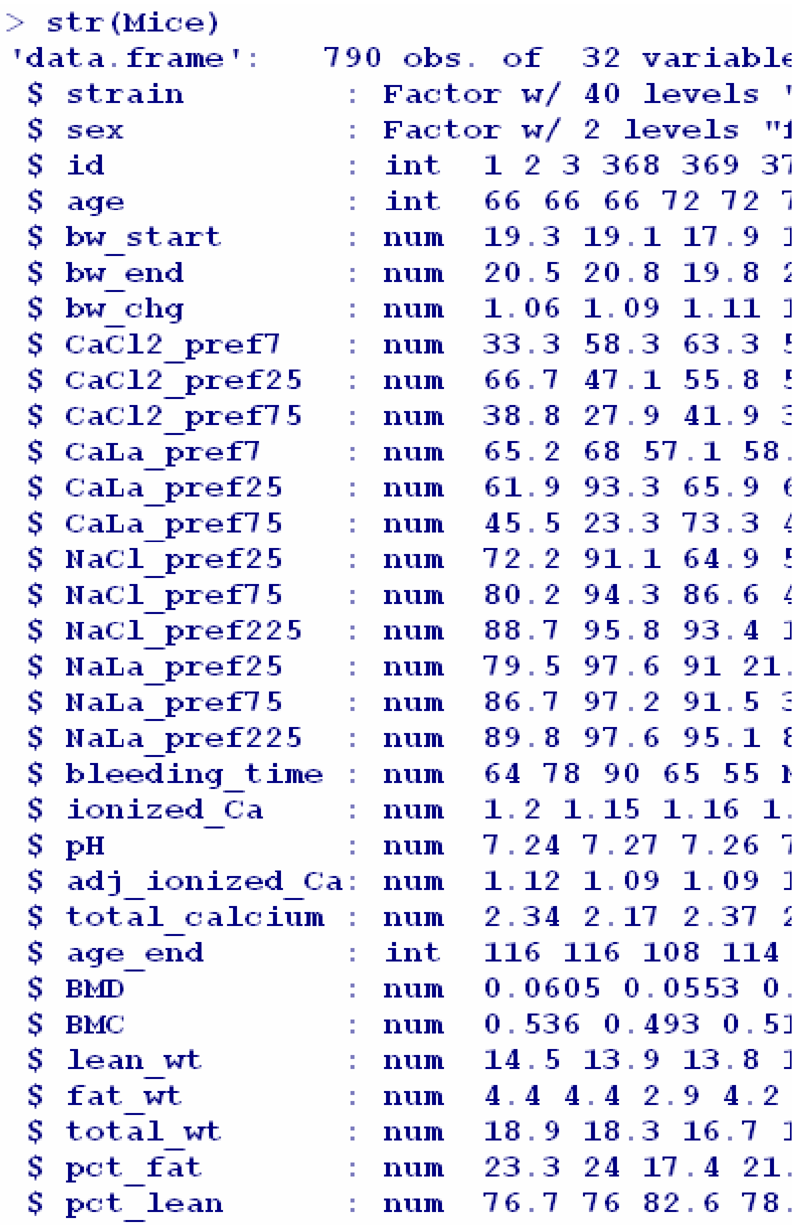
- data_for_PCA2.xls
- mice.pdf
- mice.txt
- nfl.txt
- shop.txt

6. DATA VISUALIZATION (6.3)

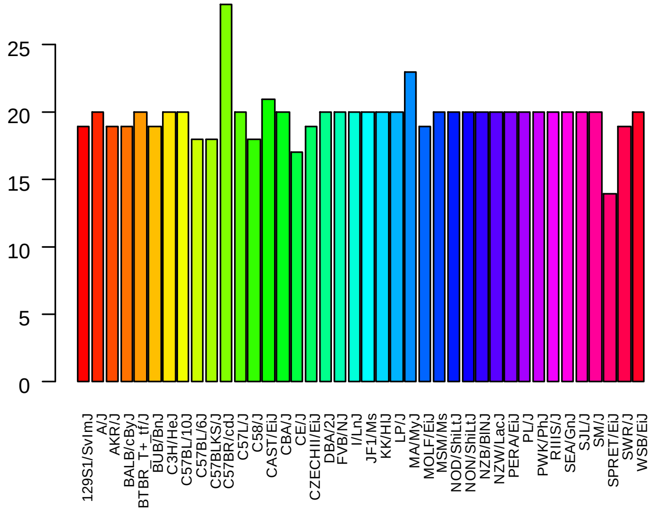
```
##-----------------------------------------------------------------
## 6.3. Mouse phenom :)
##-----------------------------------------------------------------
## load data
Mice=read.table("http://edu.sablab.net/r/data/mice.txt",
                header=T,as.is=F)
str(Mice)
## initiate window
windows(10,8)
par(mfrow=c(2,2))
## plot a factorial data
plot(Mice$strain,las=2,
     col=rainbow(nlevels(Mice$strain)),cex.names =0.7)
title("Number of mice from each strain")
## plot factorial data as pie
pie(summary(Mice$sex), col=c("pink","lightblue"))
title("Gender composition (f:female, m:male)")
## try to use special command "barplot" as well
## a histogram
hist(Mice$bw_start,probability = T,
     main="Histogram and p.d.f. approximation",
     xlab="weight, g")
lines(density(Mice$bw_start),lwd=2,col=4)

## (!) a box-plot of the population on the basis of sex
boxplot(bw_end~sex,data=Mice,col=c("pink","lightblue"))
title("Weight by sex (f:female, m:male)",
      ylab="weight, g",xlab="sex")
```



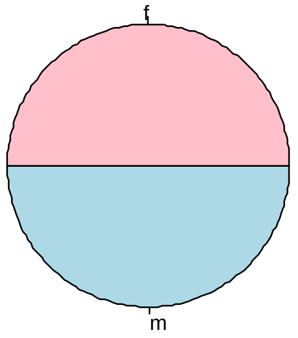
```
> str(Mice)
'data.frame':   790 obs. of  32 variable
 $ strain         : Factor w/ 40 levels '
 $ sex            : Factor w/ 2 levels "f
 $ id             : int  1 2 3 368 369 37
 $ age            : int  66 66 66 72 72 7
 $ bw_start       : num  19.3 19.1 17.9 1
 $ bw_end         : num  20.5 20.8 19.8 2
 $ bw_chg         : num  1.06 1.09 1.11 1
 $ CaCl2_pref7    : num  33.3 58.3 63.3 5
 $ CaCl2_pref25   : num  66.7 47.1 55.8 5
 $ CaCl2_pref75   : num  38.8 27.9 41.9 3
 $ CaLa_pref7     : num  65.2 68 57.1 58.
 $ CaLa_pref25    : num  61.9 93.3 65.9 6
 $ CaLa_pref75    : num  45.5 23.3 73.3 4
 $ NaCl_pref25    : num  72.2 91.1 64.9 5
 $ NaCl_pref75    : num  80.2 94.3 86.6 4
 $ NaCl_pref225   : num  88.7 95.8 93.4 1
 $ NaLa_pref25    : num  79.5 97.6 91 21.
 $ NaLa_pref75    : num  86.7 97.2 91.5 3
 $ NaLa_pref225   : num  89.8 97.6 95.1 8
 $ bleeding_time  : num  64 78 90 65 55 N
 $ ionized_Ca     : num  1.2 1.15 1.16 1.
 $ pH             : num  7.24 7.27 7.26 7
 $ adj_ionized_Ca: num  1.12 1.09 1.09 1
 $ total_calcium  : num  2.34 2.17 2.37 2
 $ age_end        : int  116 116 108 114
 $ BMD            : num  0.0605 0.0553 0.
 $ BMC            : num  0.536 0.493 0.51
 $ lean_wt        : num  14.5 13.9 13.8 1
 $ fat_wt         : num  4.4 4.4 2.9 4.2
 $ total_wt       : num  18.9 18.3 16.7 1
 $ pct_fat        : num  23.3 24 17.4 21.
 $ pct_lean       : num  76.7 76 82.6 78.
```

6. DATA VISUALIZATION (6.3)

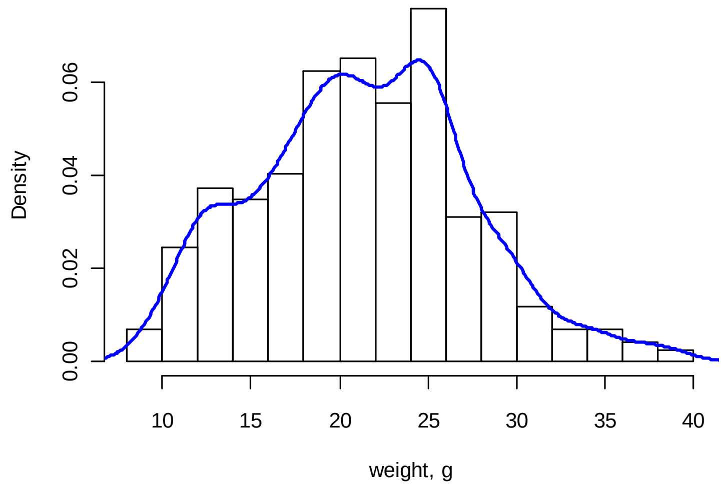
Number of mice from each strain



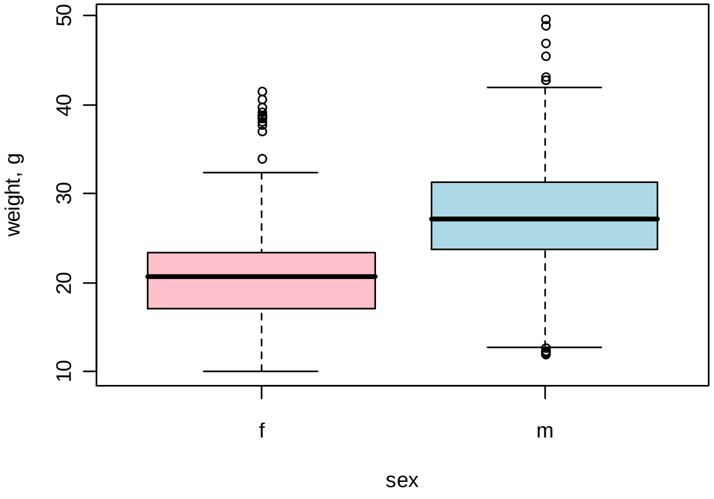
Gender composition (f:female, m:male)



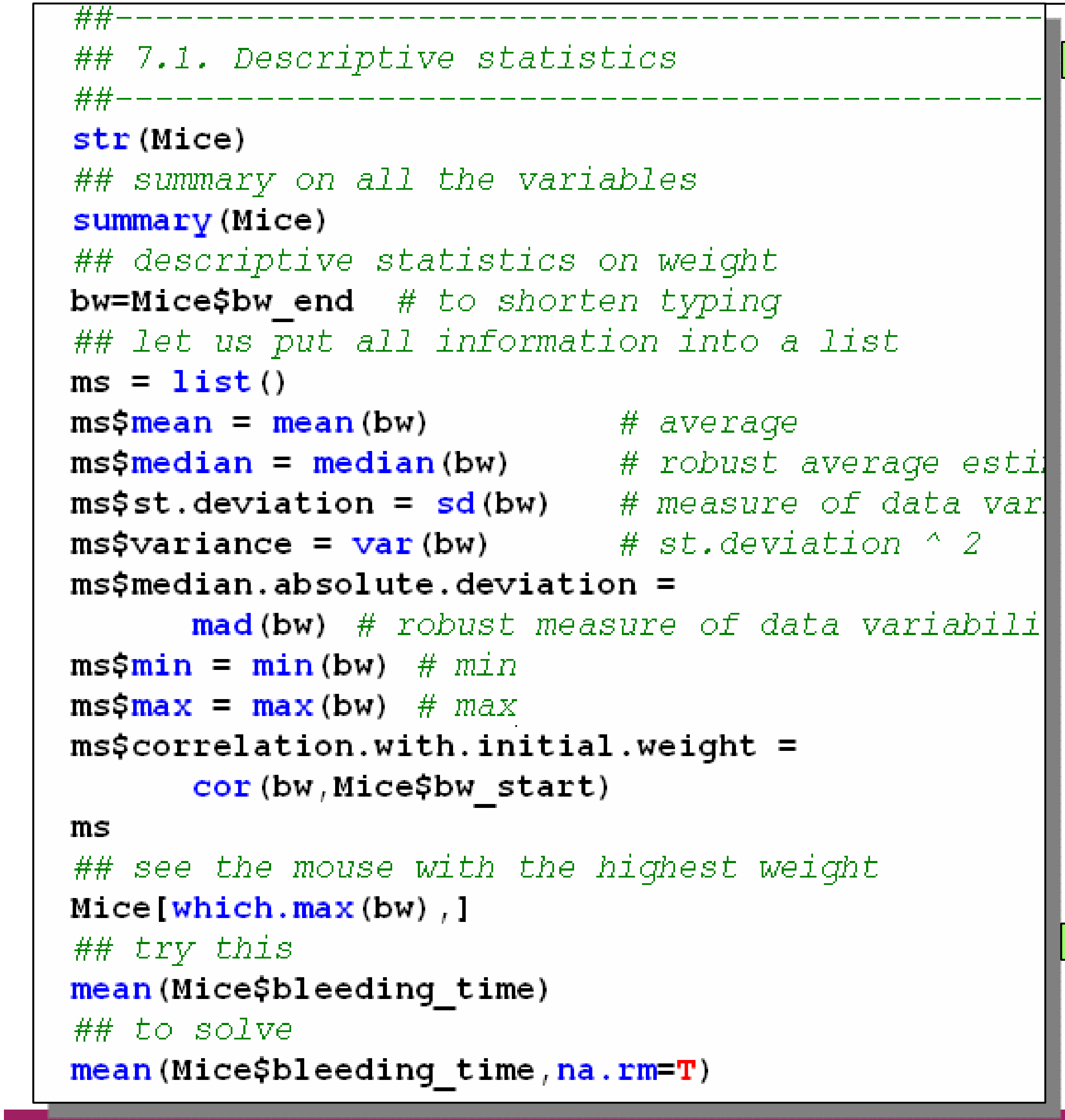
Histogram and p.d.f. approximation



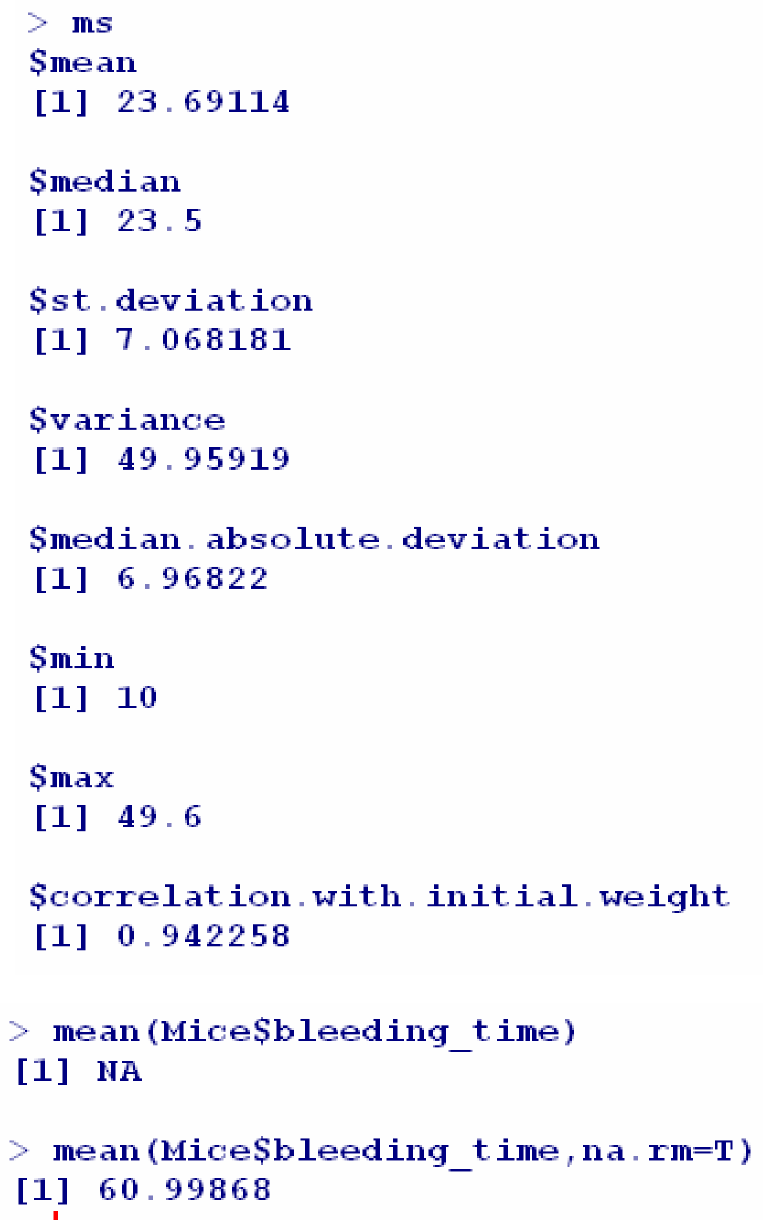
Weight by sex (f:female, m:male)



7. STATISTICAL ANALYSIS

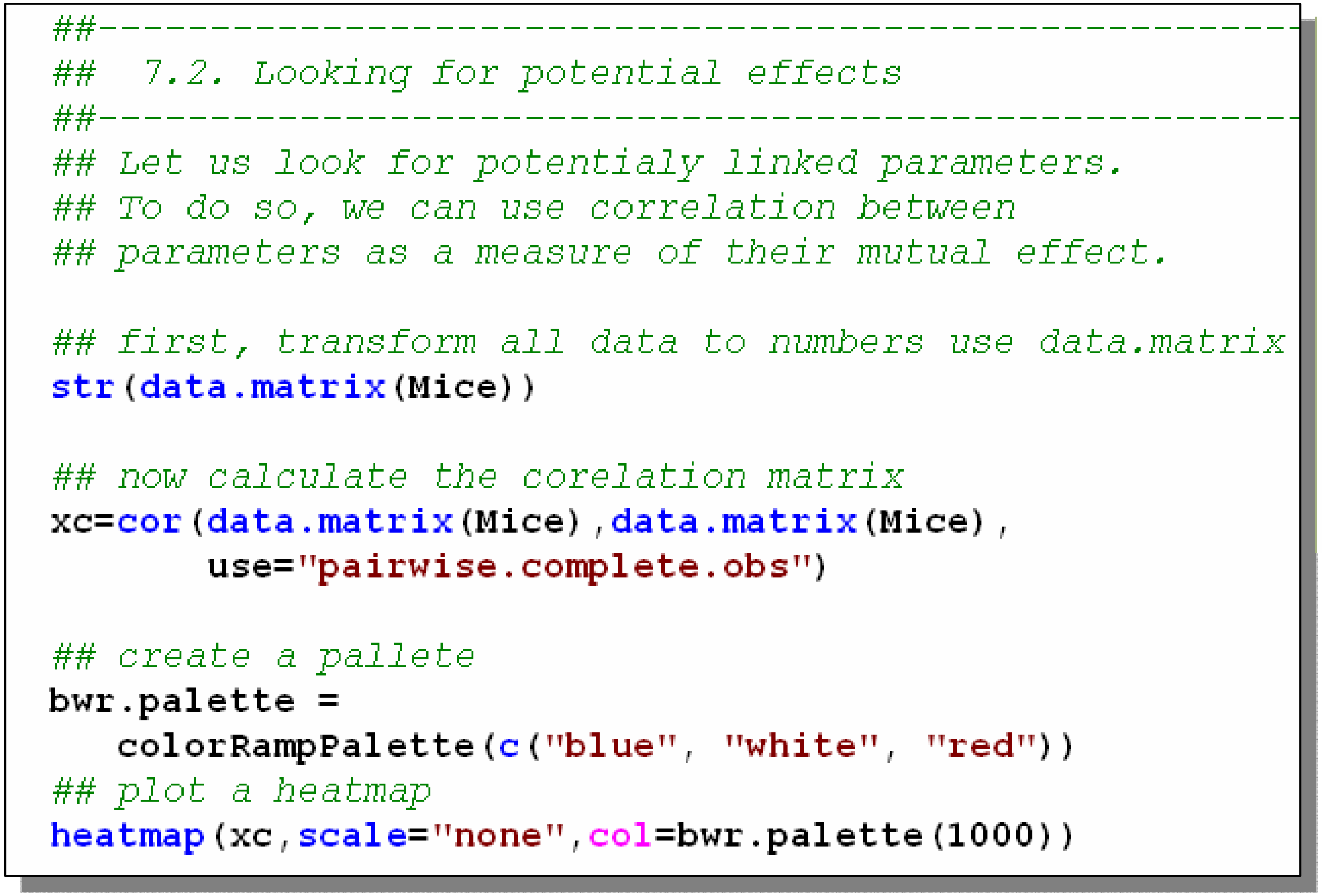


```
##---------------------------------------------------
## 7.1. Descriptive statistics
##---------------------------------------------------
str(Mice)
## summary on all the variables
summary(Mice)
## descriptive statistics on weight
bw=Mice$bw_end   # to shorten typing
## let us put all information into a list
ms = list()
ms$mean = mean(bw)             # average
ms$median = median(bw)         # robust average esti
ms$st.deviation = sd(bw)       # measure of data var
ms$variance = var(bw)          # st.deviation ^ 2
ms$median.absolute.deviation =
      mad(bw) # robust measure of data variabili
ms$min = min(bw)  # min
ms$max = max(bw)  # max
ms$correlation.with.initial.weight =
      cor(bw,Mice$bw_start)
ms
## see the mouse with the highest weight
Mice[which.max(bw),]
## try this
mean(Mice$bleeding_time)
## to solve
mean(Mice$bleeding_time,na.rm=T)
```

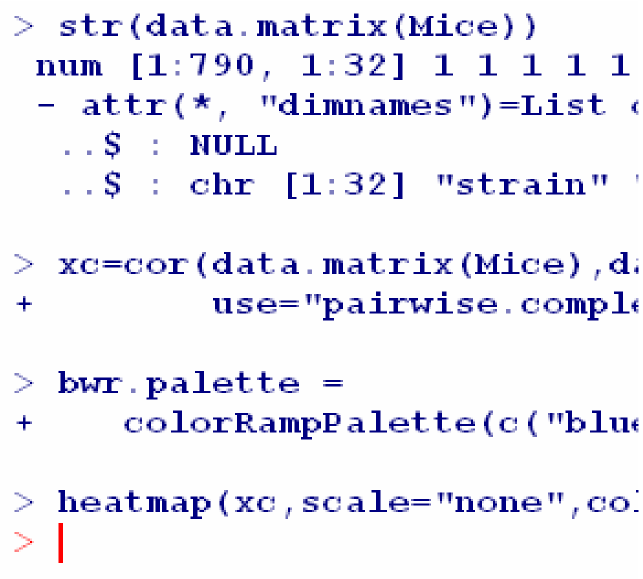


```
> ms
$mean
[1] 23.69114

$median
[1] 23.5

$st.deviation
[1] 7.068181

$variance
[1] 49.95919

$median.absolute.deviation
[1] 6.96822

$min
[1] 10

$max
[1] 49.6

$correlation.with.initial.weight
[1] 0.942258


> mean(Mice$bleeding_time)
[1] NA

> mean(Mice$bleeding_time,na.rm=T)
[1] 60.99868
```

7. STATISTICAL ANALYSIS

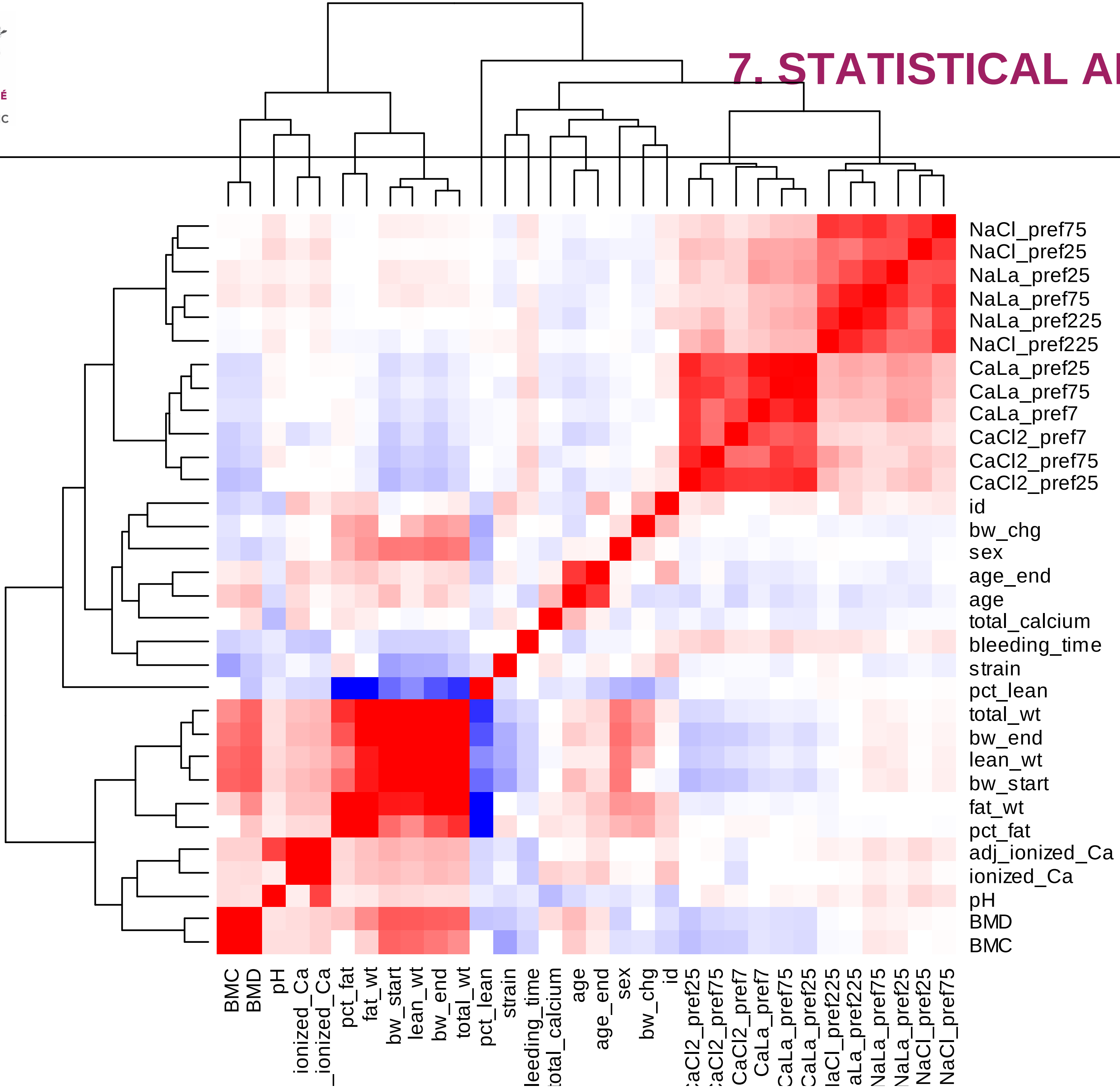


```
##-------------------------------------------------------------
##  7.2. Looking for potential effects
##-------------------------------------------------------------
## Let us look for potentialy linked parameters.
## To do so, we can use correlation between
## parameters as a measure of their mutual effect.

## first, transform all data to numbers use data.matrix
str(data.matrix(Mice))

## now calculate the corelation matrix
xc=cor(data.matrix(Mice),data.matrix(Mice),
       use="pairwise.complete.obs")

## create a pallete
bwr.palette =
   colorRampPalette(c("blue", "white", "red"))
## plot a heatmap
heatmap(xc,scale="none",col=bwr.palette(1000))
```



```
> str(data.matrix(Mice))
 num [1:790, 1:32] 1 1 1 1 1
 - attr(*, "dimnames")=List 
  ..$ : NULL
  ..$ : chr [1:32] "strain" 

> xc=cor(data.matrix(Mice),d
+        use="pairwise.comple

> bwr.palette =
+    colorRampPalette(c("blue

> heatmap(xc,scale="none",co
> 
```

7. STATISTICAL ANALYSIS



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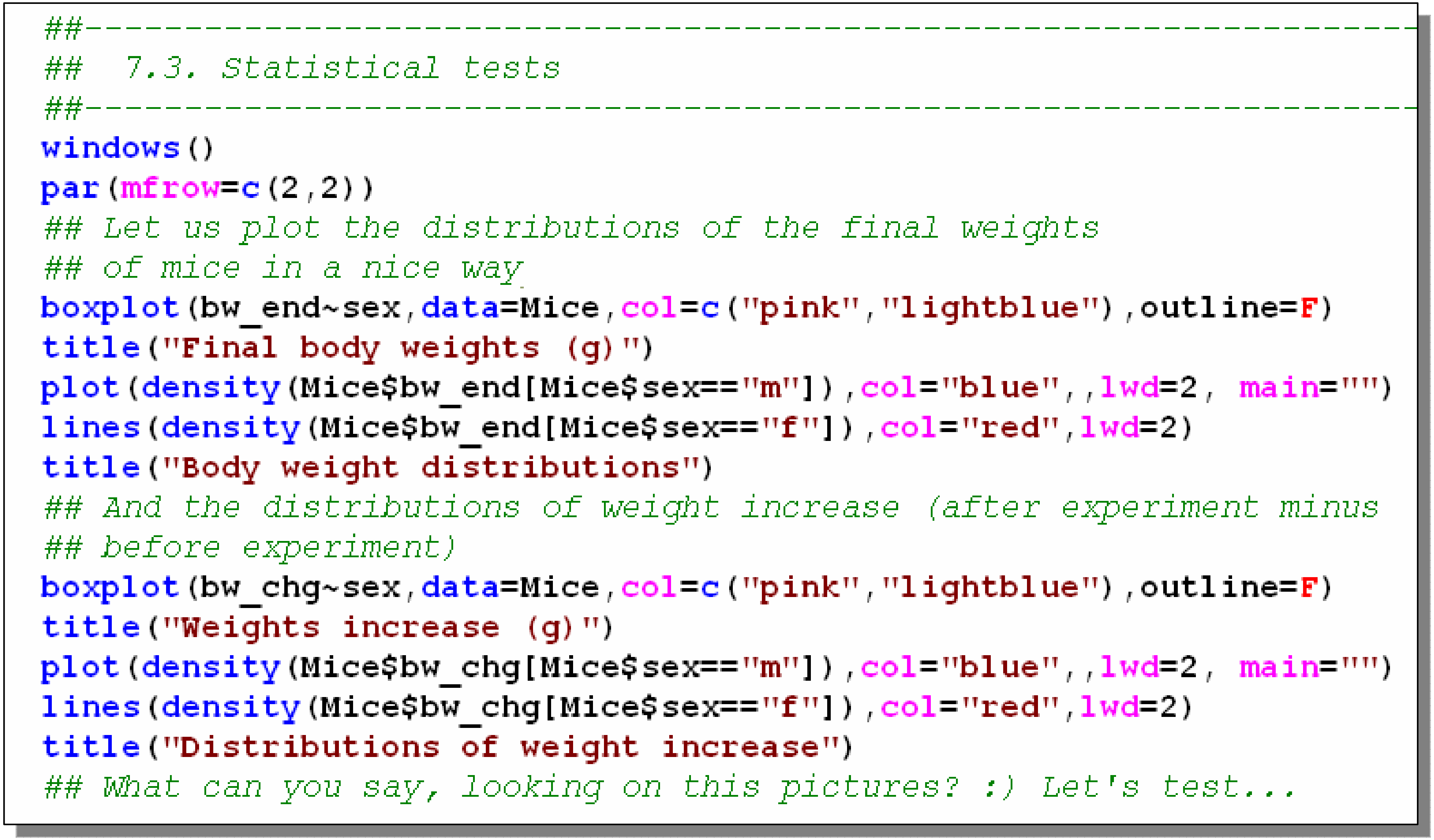
7. STATISTICAL ANALYSIS



```
##-------------------------------------------------------------------
##  7.3. Statistical tests
##-------------------------------------------------------------------
windows()
par(mfrow=c(2,2))
## Let us plot the distributions of the final weights
## of mice in a nice way
boxplot(bw_end~sex,data=Mice,col=c("pink","lightblue"),outline=F)
title("Final body weights (g)")
plot(density(Mice$bw_end[Mice$sex=="m"]),col="blue",,lwd=2, main="")
lines(density(Mice$bw_end[Mice$sex=="f"]),col="red",lwd=2)
title("Body weight distributions")
## And the distributions of weight increase (after experiment minus
## before experiment)
boxplot(bw_chg~sex,data=Mice,col=c("pink","lightblue"),outline=F)
title("Weights increase (g)")
plot(density(Mice$bw_chg[Mice$sex=="m"]),col="blue",,lwd=2, main="")
lines(density(Mice$bw_chg[Mice$sex=="f"]),col="red",lwd=2)
title("Distributions of weight increase")
## What can you say, looking on this pictures? :) Let's test...
```

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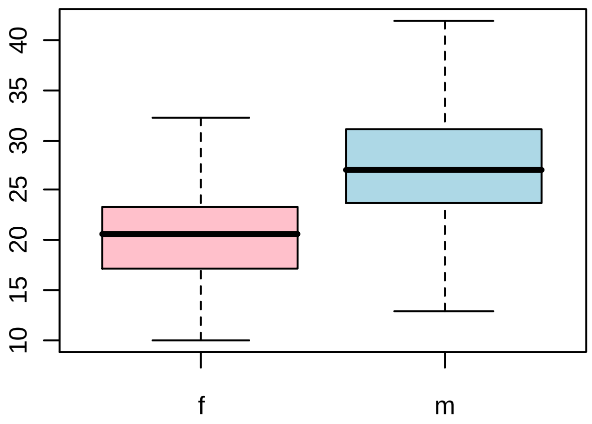
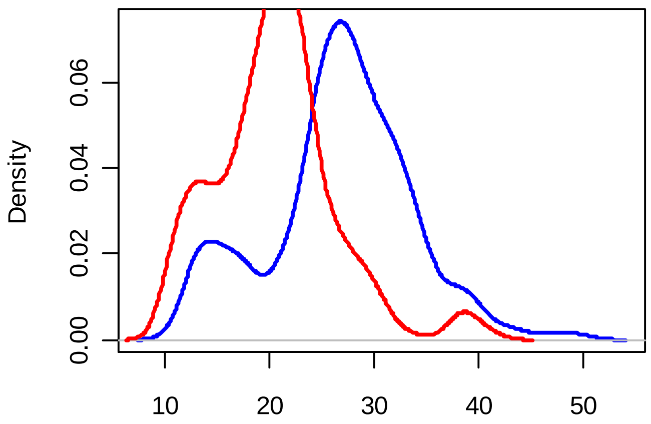
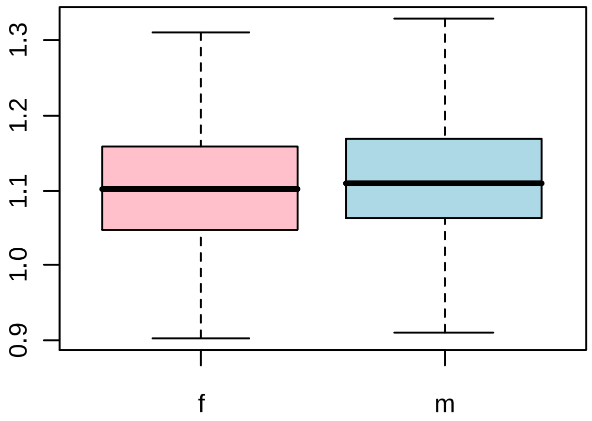
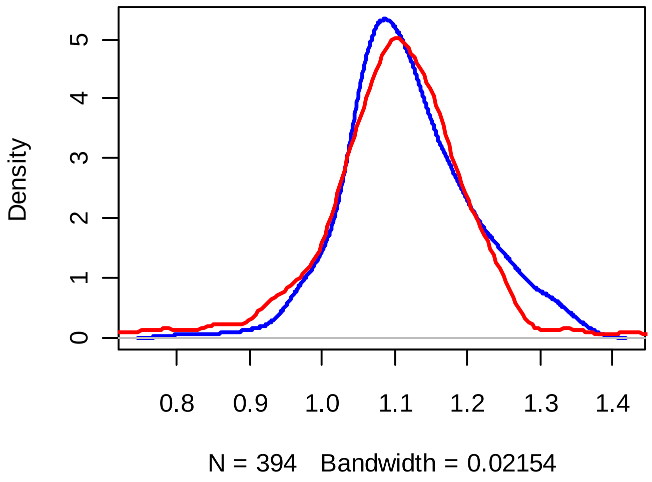
7. STATISTICAL ANALYSIS


Final body weights (g)


Body weight distributions

What can you say looking on these plots?


Weights increase (g)


Distributions of weight increase

7. STATISTICAL ANALYSIS

Well, in fact almost nothing…

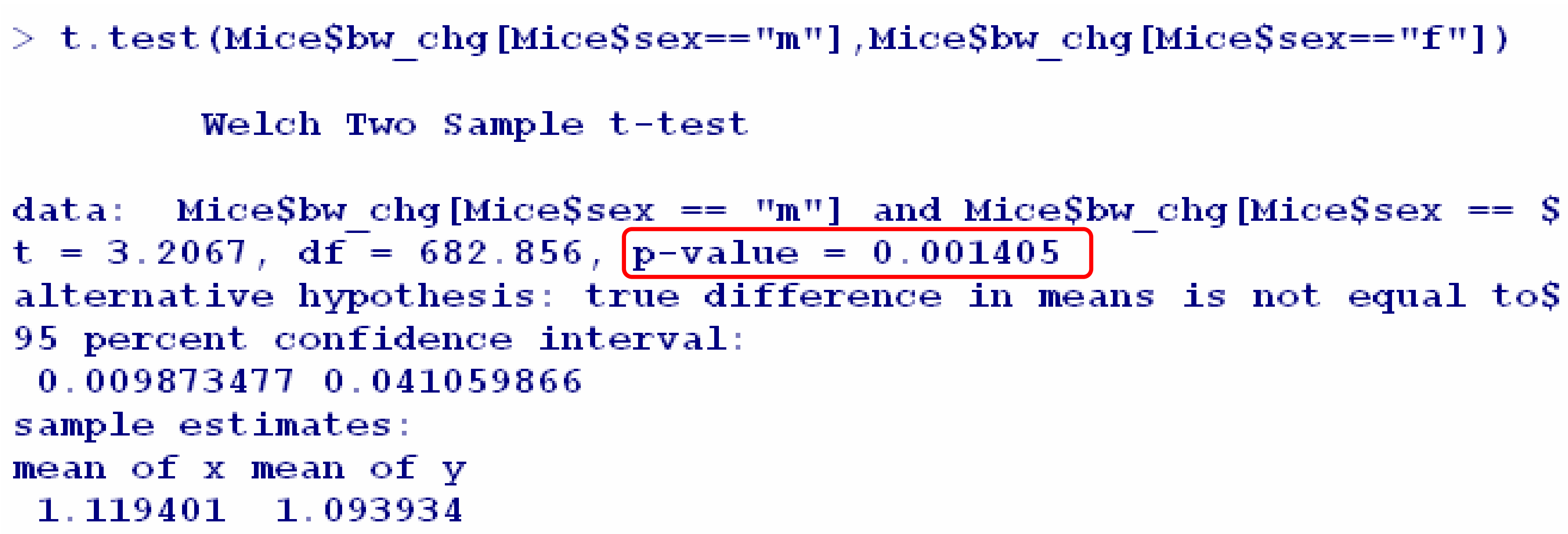
```
## Perform t.tests (by default - unpaired, two.sided)
t.test(Mice$bw_end[Mice$sex=="m"],Mice$bw_end[Mice$sex=="f"])
t.test(Mice$bw_chg[Mice$sex=="m"],Mice$bw_chg[Mice$sex=="f"])

## Perform Wilcoxon Rank Sum (equiv. Mann-Whitney test)
wilcox.test(Mice$bw_end[Mice$sex=="m"],Mice$bw_end[Mice$sex=="f"])
wilcox.test(Mice$bw_chg[Mice$sex=="m"],Mice$bw_chg[Mice$sex=="f"])
```

p-value = 2.2e-16

p-value = 0.0014

p-value = 2.2e-16

p-value = 0.029

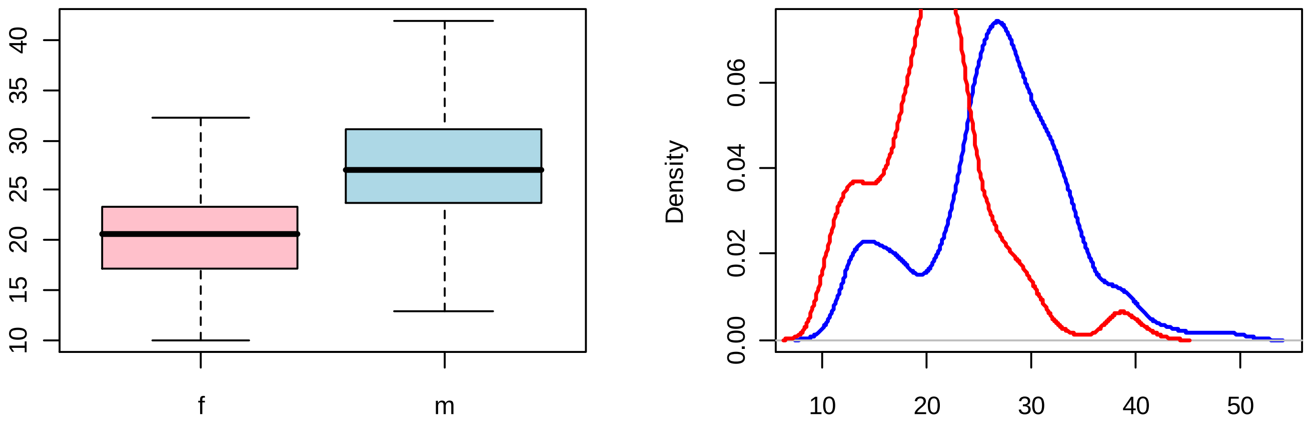


```
> t.test(Mice$bw_chg[Mice$sex=="m"],Mice$bw_chg[Mice$sex=="f"])

        Welch Two Sample t-test

data:  Mice$bw_chg[Mice$sex == "m"] and Mice$bw_chg[Mice$sex == $
t = 3.2067, df = 682.856, p-value = 0.001405
alternative hypothesis: true difference in means is not equal to$
95 percent confidence interval:
 0.009873477 0.041059866
sample estimates:
mean of x mean of y
 1.119401  1.093934
```

7. STATISTICAL ANALYSIS

Final body weights (g)

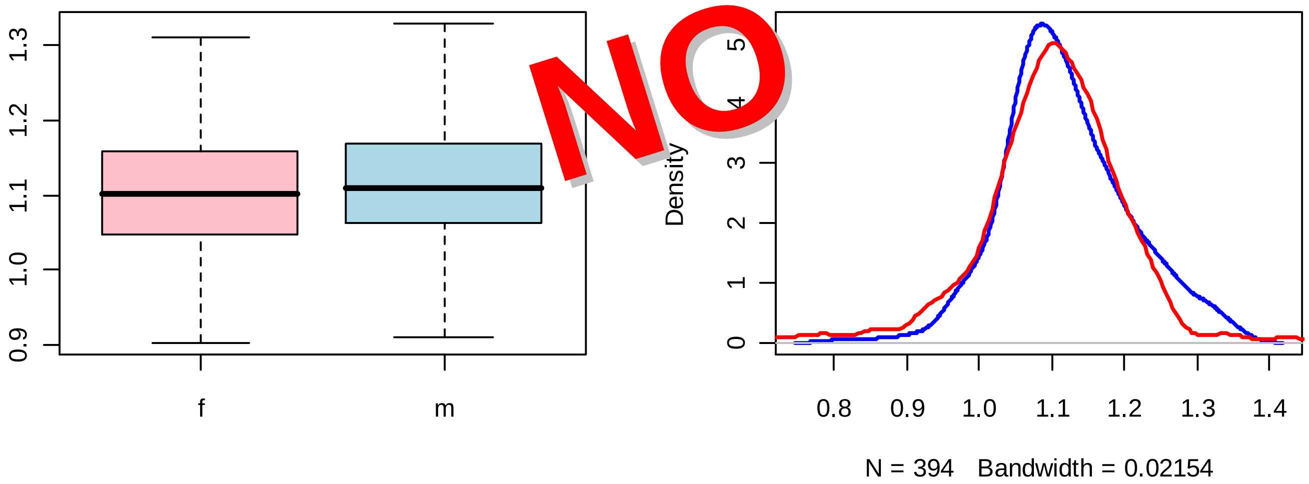
Body weight distributions



By the way, can you distinguish M and F by weight?

Weights increase (g)

Distributions of weight increase



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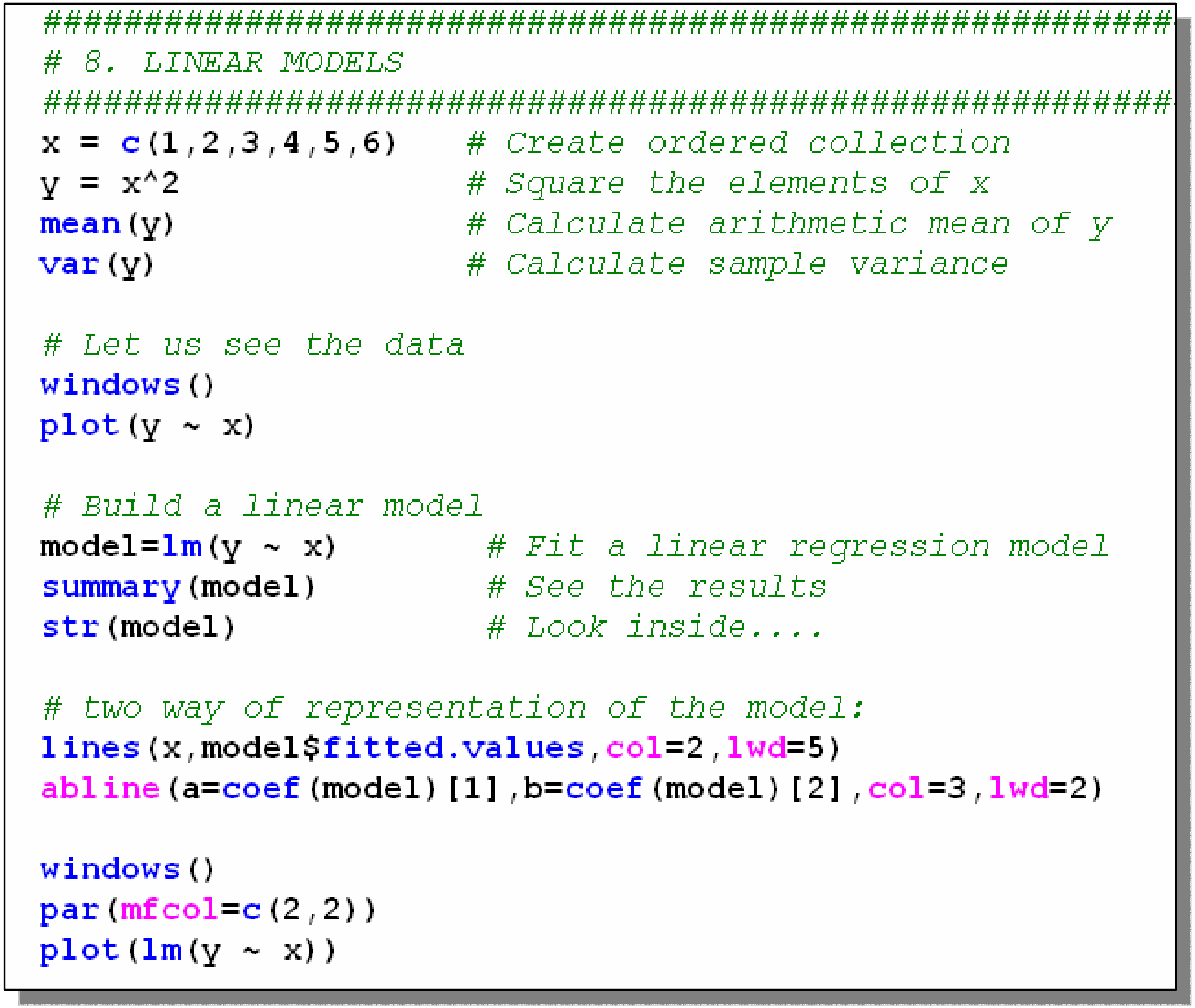
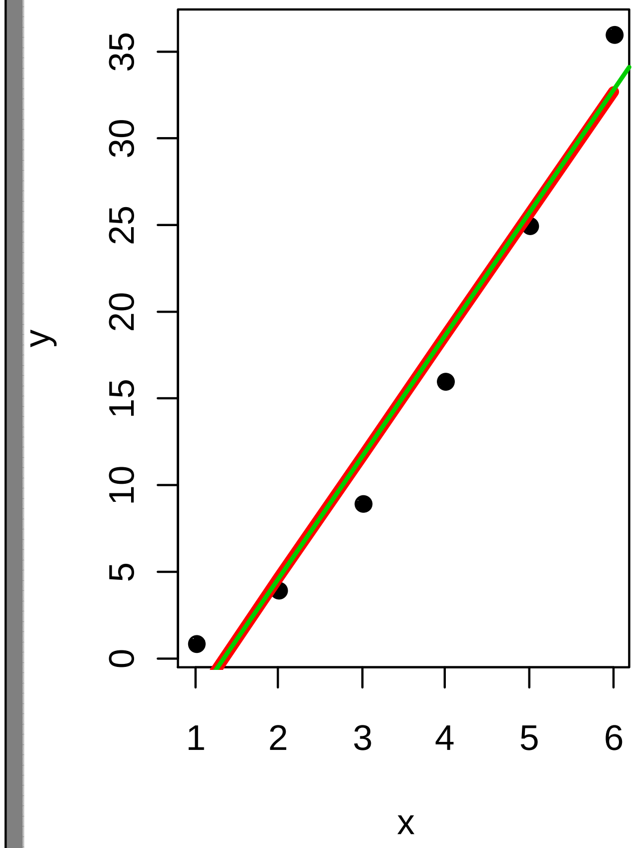
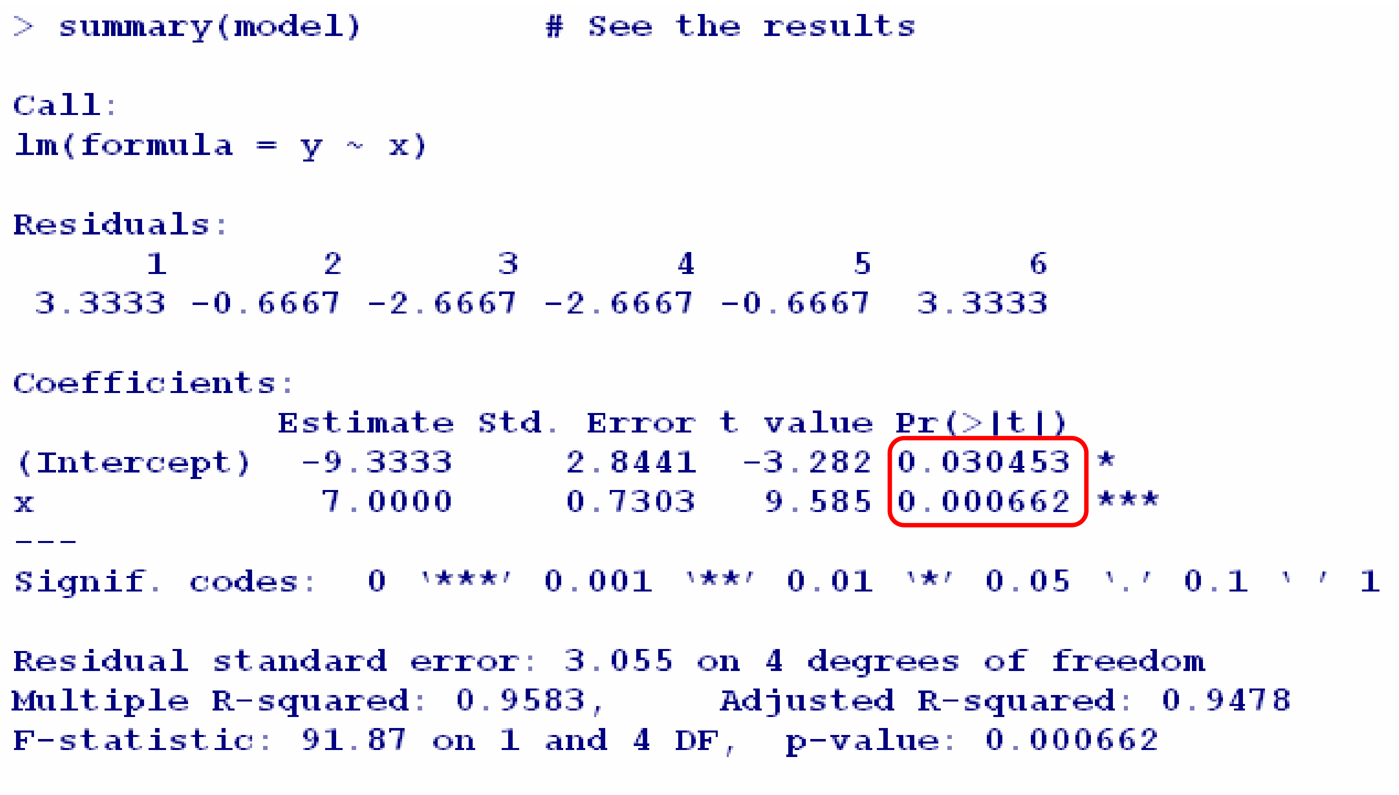
8. MODEL FITTING



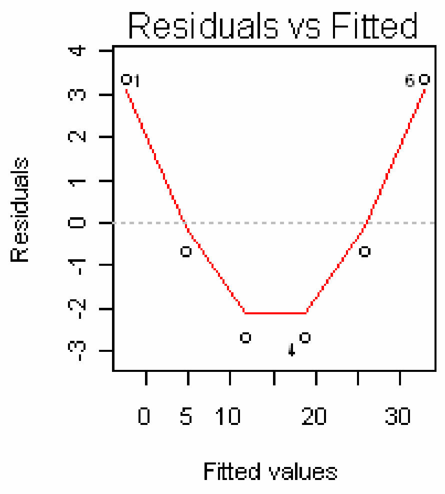
```
#########################################################
# 8. LINEAR MODELS
#########################################################
x = c(1,2,3,4,5,6)    # Create ordered collection
y = x^2               # Square the elements of x
mean(y)               # Calculate arithmetic mean of y
var(y)                # Calculate sample variance

# Let us see the data
windows()
plot(y ~ x)

# Build a linear model
model=lm(y ~ x)        # Fit a linear regression model
summary(model)         # See the results
str(model)             # Look inside....

# two way of representation of the model:
lines(x,model$fitted.values,col=2,lwd=5)
abline(a=coef(model)[1],b=coef(model)[2],col=3,lwd=2)

windows()
par(mfcol=c(2,2))
plot(lm(y ~ x))
```

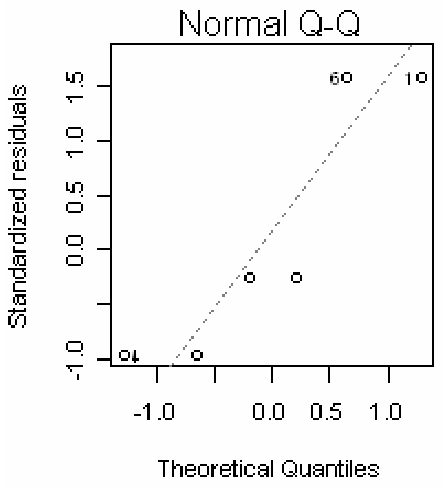


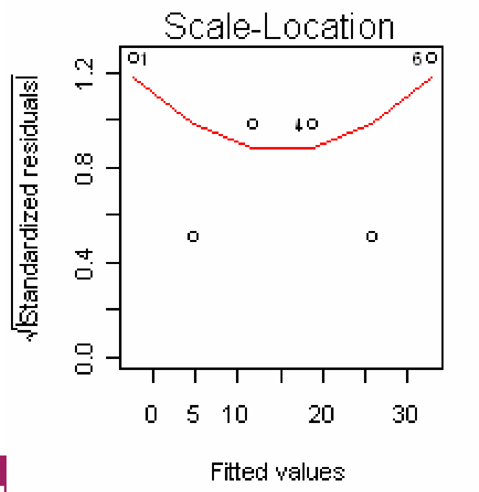
8. MODEL FITTING

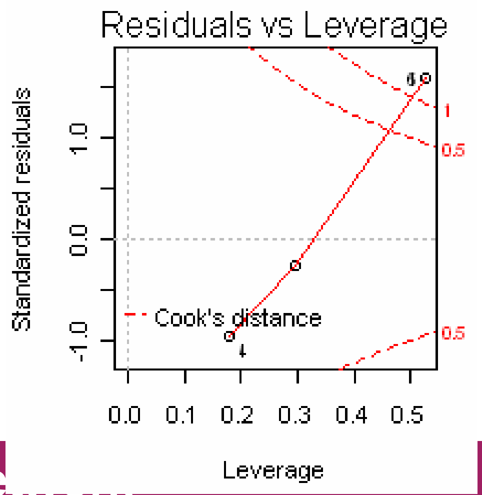


```
> summary(model)        # See the results

Call:
lm(formula = y ~ x)

Residuals:
      1       2       3       4       5       6
 3.3333 -0.6667 -2.6667 -2.6667 -0.6667  3.3333

Coefficients:
            Estimate Std. Error t value Pr(>|t|)
(Intercept)  -9.3333     2.8441  -3.282 0.030453 *
x             7.0000     0.7303   9.585 0.000662 ***
---
Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

Residual standard error: 3.055 on 4 degrees of freedom
Multiple R-squared: 0.9583,     Adjusted R-squared: 0.9478
F-statistic: 91.87 on 1 and 4 DF,  p-value: 0.000662
```

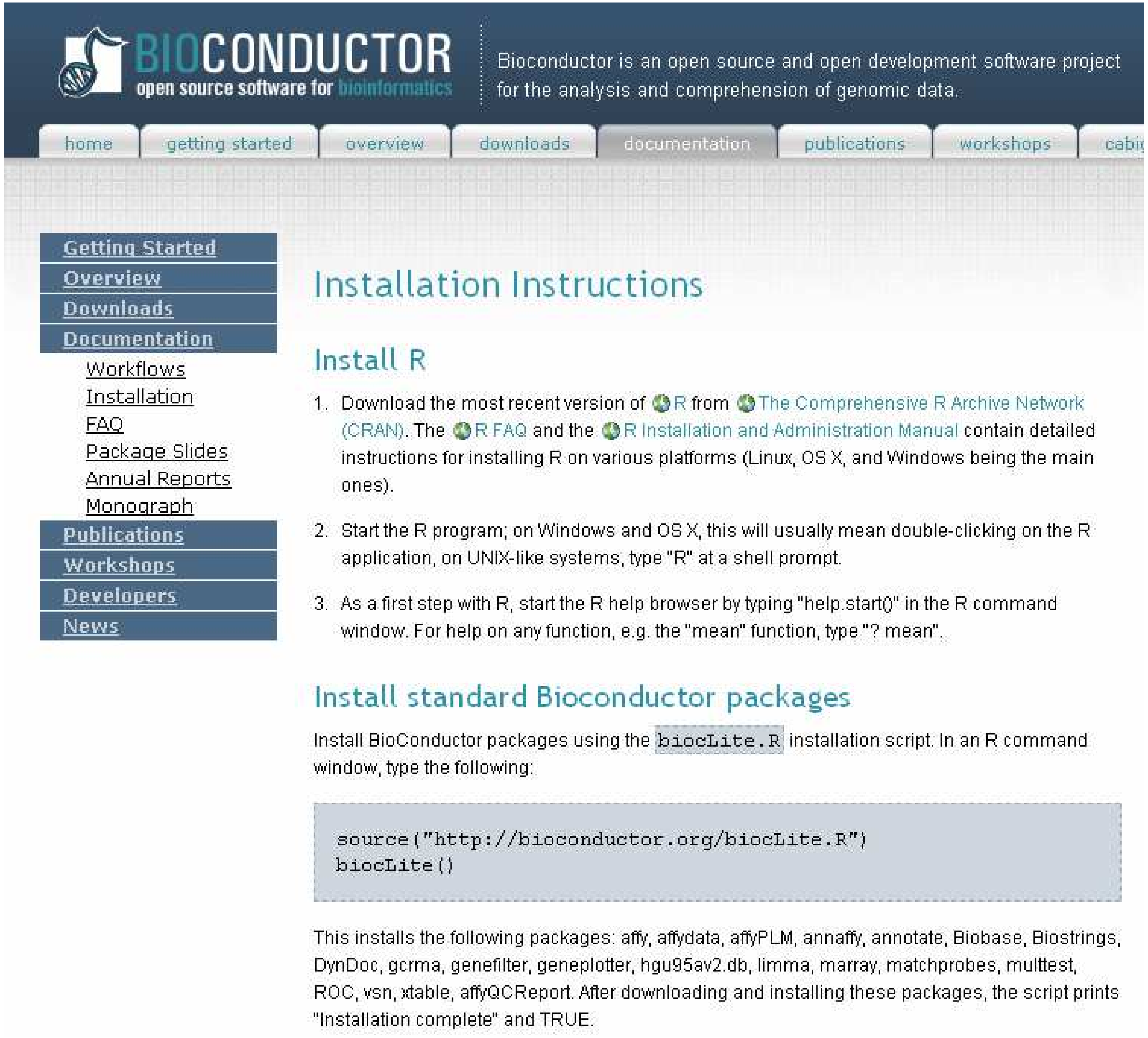








9. BIOCONDUCTOR



Bioconductor is an open source and open development software project for the analysis and comprehension of genomic data.

Installation Instructions

Install R

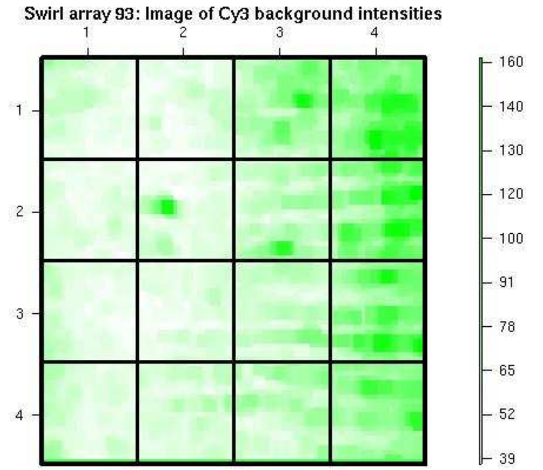
1. Download the most recent version of R from The Comprehensive R Archive Network (CRAN). The R FAQ and the R Installation and Administration Manual contain detailed instructions for installing R on various platforms (Linux, OS X, and Windows being the main ones).
2. Start the R program; on Windows and OS X, this will usually mean double-clicking on the R application, on UNIX-like systems, type "R" at a shell prompt.
3. As a first step with R, start the R help browser by typing "help.start()" in the R command window. For help on any function, e.g. the "mean" function, type "? mean".

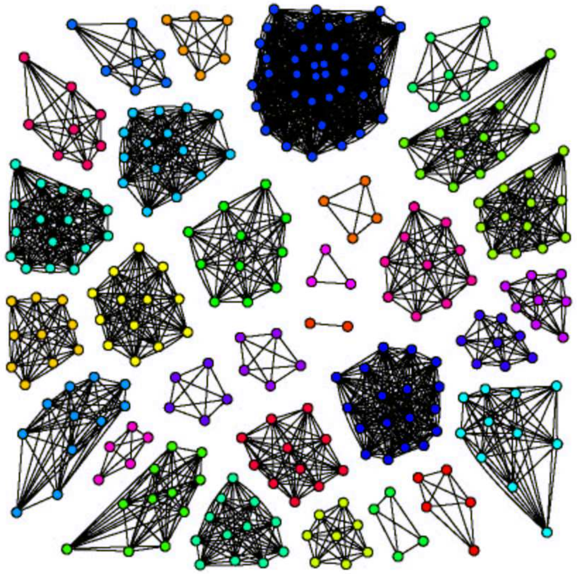
Install standard Bioconductor packages

Install BioConductor packages using the `biocLite.R` installation script. In an R command window, type the following:

```
source("http://bioconductor.org/biocLite.R")
biocLite()
```

This installs the following packages: affy, affydata, affyPLM, annaffy, annotate, Biobase, Biostrings, DynDoc, gcrma, genefilter, geneplotter, hgu95av2.db, limma, marray, matchprobes, multtest, ROC, vsn, xtable, affyQCReport. After downloading and installing these packages, the script prints "Installation complete" and TRUE.





www.bioconductor.org

Thank you for your attention

to be continued…