

Swiss Institute of
Bioinformatics

First steps with R in Life Sciences: Introduction

February, 2022 - Streamed

Wandrille Duchemin

-- with slides from Diana Marek, Wandrille Duchemin, Leonore Wigger



www.sib.swiss

General info

Schedule:

09:00 – 17:00

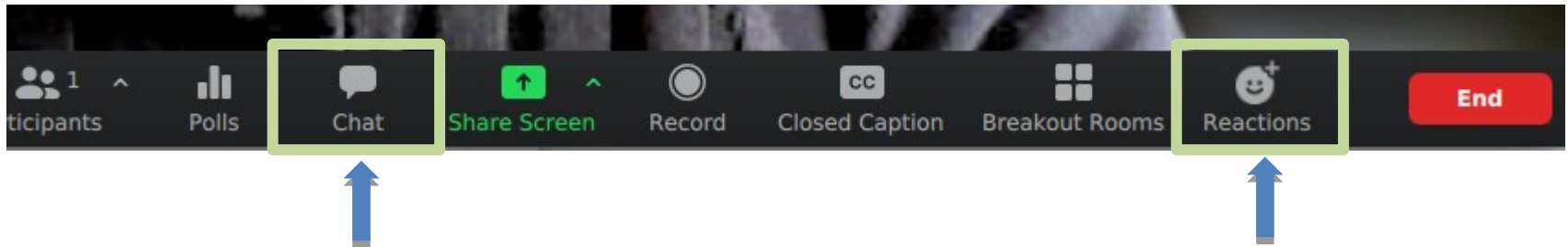
coffee-breaks : ~10:15 & ~15:15

lunch break : 12:00 – 13:00

Course Material (Slides, Data Sets, Exercises) available on github.

Optional exam, 0.5 ECTS value

Zoom communication



Please rename yourself to “name surname”
Do not hesitate to ask questions in the chat
(Helpers are here to answer!)

Lectures : please mute yourselves (raise-hand for questions)

Exercise sessions :

do not hesitate to open your mic and webcam
so we may communicate

Introducing ourselves


In the google Doc

- Write your name and 3 keywords that defines your activity and/or interest in R
- Additional questions about your previous experience and interest for the topics

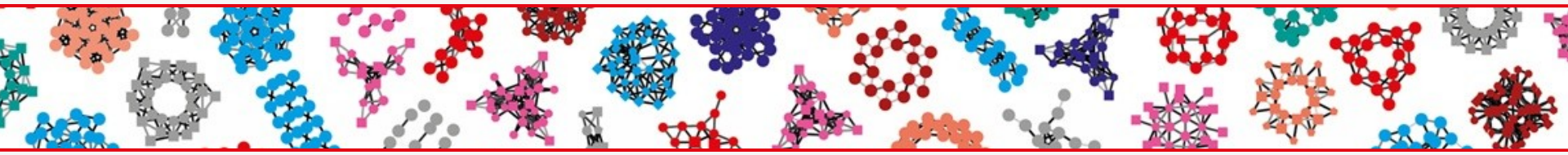
Choice of content

R is extremely vast and can't be learned overnight. The scope of this course is to:

- give a **basic understanding and concepts behind R**
- help implement and interpret a data analysis workflow

This course is only the first step in your  journey

Outline



Day 1

01

What is R? Introduction

02

Getting familiar with R and the RStudio environment

03

Getting started with R syntax and objects

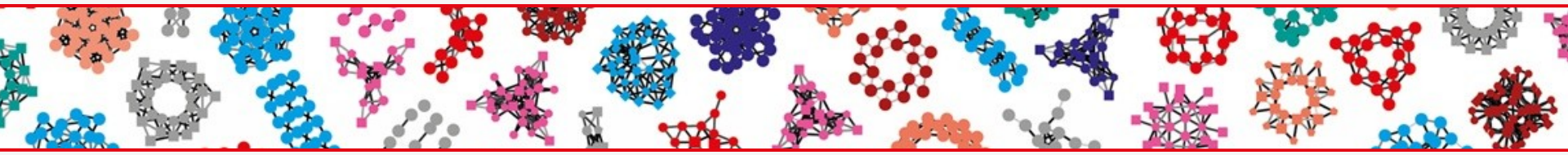
04

Formatting your data

05

Importing/exporting data with R

Outline



Day 2

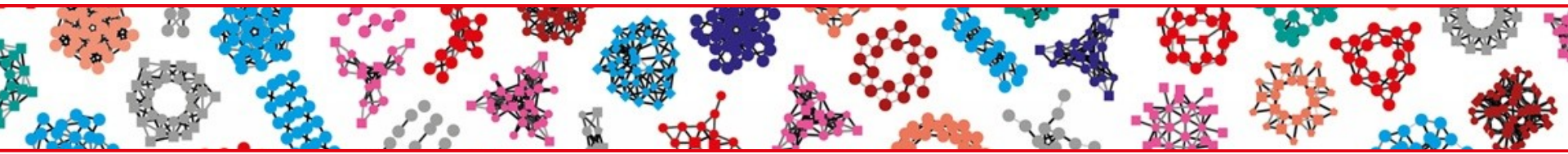
06

Building graphics in R (basic plotting)

07

Starting with statistics in R (hypothesis testing, simple linear regression)

Examples and exercises are integrated in the chapters



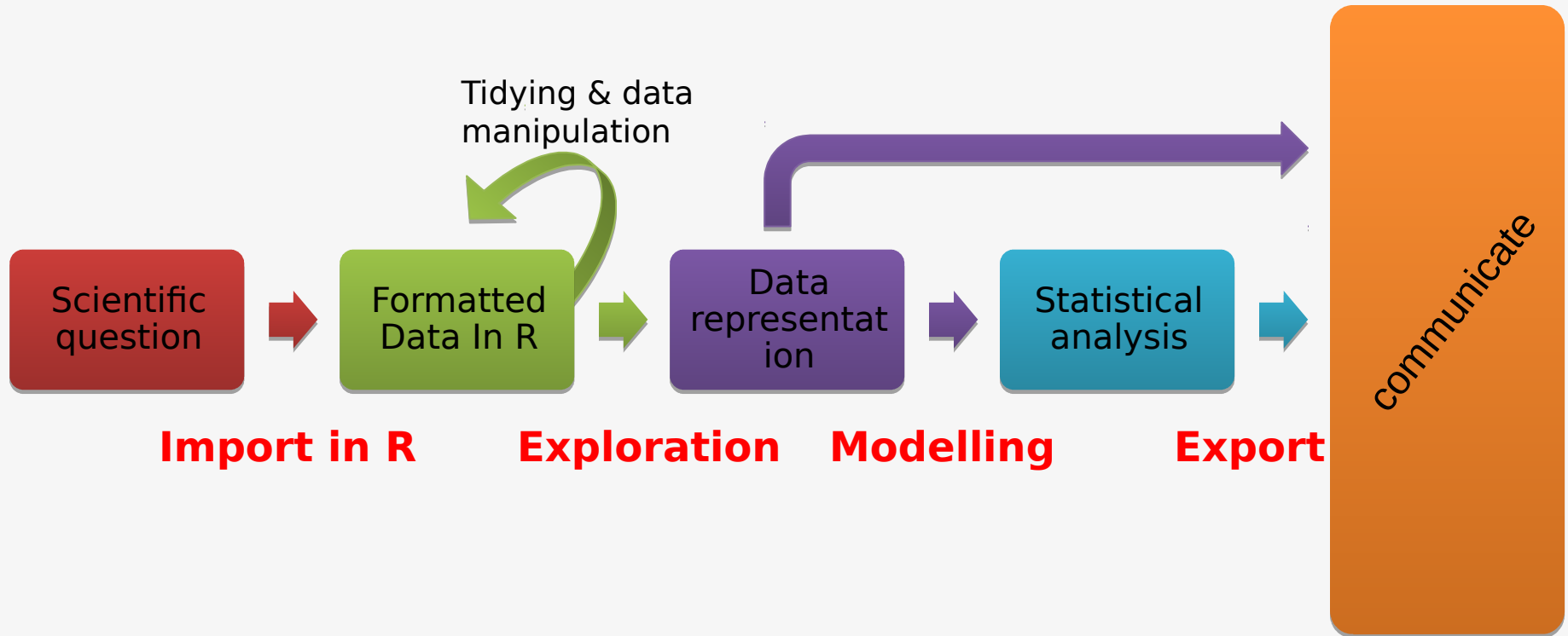
01

What is R? Introduction

What is R?

- R is a programming language and an environment for statistical computing and graphics.
- A simple development environment with a console and a text editor
- Facilities for data import, storage and manipulation
- Functions for calculations on vectors and matrices
- Large collections of data analysis tools
- Graphical tools

Taking advantage of R for your work

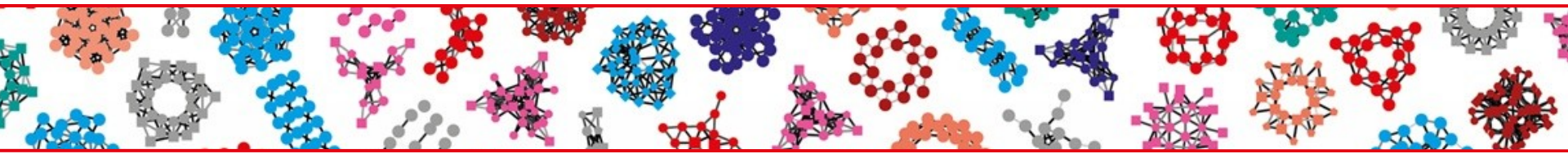


R's success

- R has become the **tool of choice** for statistical analysis in several fields, including **life sciences** and finance.
- Why?
 - It's free
 - It's well documented
 - Large number of contributed packages are freely available, easy to install, for nearly every type of statistics, machine learning, high-quality plots

R's user community

- Group of **core developers** who **maintain** and **upgrade** the basic R installation. New version every 6 months.
- Anyone can contribute with **add-on packages** which provide additional functionality (thousands of such packages available).
- **Online help**
 - in user group forums, eg:
<https://stat.ethz.ch/mailman/listinfo/r-help>
<http://stackoverflow.com/questions/tagged/r>
 - in countless online tutorials, books, blogs



02

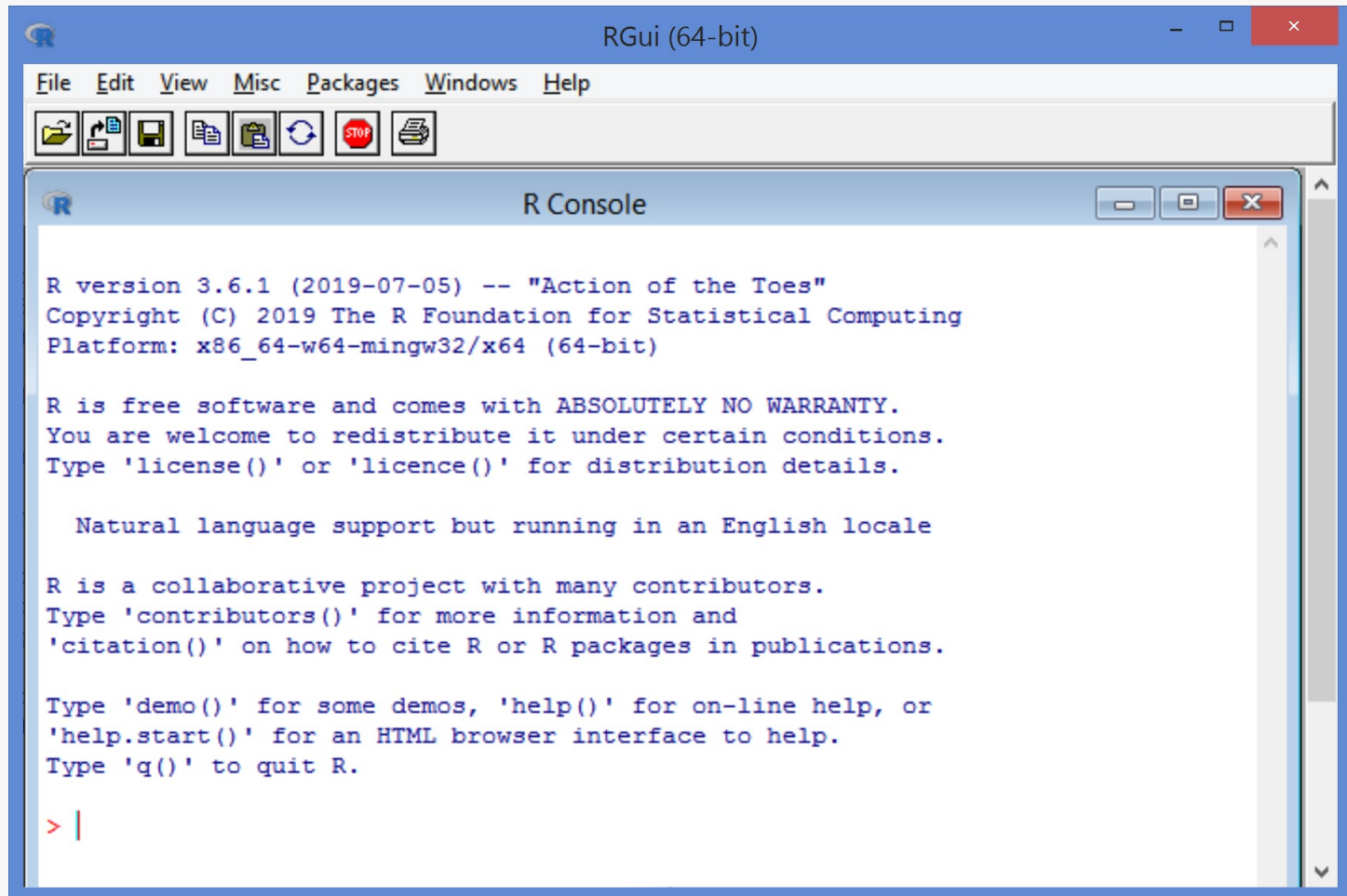
Getting familiar with R and RStudio environments

R installation

- R can be freely downloaded, copied, used, modified or redistributed.
- From the main website <http://www.r-project.org> choose "CRAN", "Download", then pick a mirror website close to you (e.g. in Switzerland).
- You can then download and install the correct version for your operating system (Windows, Linux, Mac).

RGui (R Graphical user interface) - P5S

- Together with the programming language, a graphical user interface is installed.



R combined with RStudio

<http://www.rstudio.com/>

RStudio is an integrated development environment (IDE),
designed to help you be more productive with R

It includes:

- A console
- A syntax-highlighting editor that supports direct code execution
- Tools for viewing the workspace and the history
- A file explorer, a package explorer, plot and help display

We suggest Rstudio as a more powerful, more comfortable alternative to the RGUI

RStudio interface

The screenshot shows the RStudio interface with the following components labeled:

- Editor**: The central area for writing R code. It contains a script named `first_script.R` with the following content:

```
1 ##### My first script #####
2 ##### October 2017 #####
3
4
5 # list workspace
6 ls()
7
8 # Reset R's brain
9 rm(list=ls())
10
11 # check wd
12 getwd()
13
14 #set wd
15 setwd("~/Users/dmarek/EducationSIB/Courses_2016/First_Steps_R_June2016/R_intro_course")
16
17 # confirm wd
18 getwd()
19
20 #load packages if needed (to do every time you launch your R session)
21 #library("boot")
22 #library("lattice")
23
24 ##### October 2017 #####
```
- Workspace, history**: The **Environment** pane on the right, showing the current workspace. It lists objects: `mice_data` (50 obs. of 3 variables), `mice_weight_HFD` (29 obs. of 3 variables), and `mids` (num [1:2, 1] 0.7 1.9). Below this, the **Values** pane shows summary statistics for `mean_weight_diet`, `mean_weight_genotype`, `n_weight_diet`, `n_weight_genotype`, `sd_weight_diet`, and `sd_weight_genotype`.
- Console, terminal**: The **Console** pane at the bottom left, showing the R startup message and instructions on how to use R. The text is:

R est un logiciel libre livré sans AUCUNE GARANTIE. Vous pouvez le redistribuer sous certaines conditions. Tapez 'license()' ou 'licence()' pour plus de détails.

R est un projet collaboratif avec de nombreux contributeurs. Tapez 'contributors()' pour plus d'information et 'citation()' pour la façon de le citer dans les publications.

Tapez 'demo()' pour des démonstrations, 'help()' pour l'aide en ligne ou 'help.start()' pour obtenir l'aide au format HTML. Tapez 'q()' pour quitter R.

[Workspace loaded from ~/TrainingSIB/Courses_2017/First_Steps_R_Oct2017/.RData]
- File explorer, plots, packages, help**: The **Files** pane on the bottom right, showing a file explorer view of the `course_datasets` directory. It lists files such as `class.csv`, `etubiol.csv`, `mammals_survey.csv`, `melanoma_data.txt`, `mice_data.csv`, `my_data_frame.csv`, `pigs.csv`, `smoker.csv`, and `snp.csv` with their respective sizes and modification dates.

Creating an R project

- RStudio allows organizing your work into projects.
 - Go to **File > New project** or click on “Project” in the upper right corner of RStudio.
 - Choose **New Directory**, then **New Project**, give a name to the directory and set its location.
 - This **creates a new directory which contains a .Rproj file** (same name as the directory).
 - OR choose **Existing Directory**, click **Browse**, navigate to a folder, then click **Create Project**
 - This **creates an .Rproj file inside the directory** (same name as the directory).

The project directory becomes automatically the working directory.

This is one of the ways RStudio adds convenience

Let's practice – 1

- 1) On your computer, create a folder for this course.
- 2) Download the `course_datasets.zip` file, **unzip it** in the course folder.
- 3) In RStudio, create a new project in the course folder.

Console: The Command Line

~/TrainingSIB/Courses_2017/First_Steps_R_Oct2017/ ↗

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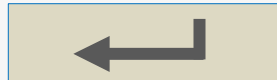
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en ligne ou 'help.start()' pour obtenir l'aide au format HTML.
Tapez 'q()' pour quitter R.

[Workspace loaded from ~/TrainingSIB/Courses_2017/First_Steps_R_Oct2017/.RData]

> |

**The prompt ">" indicates that R
is waiting for you to type a
command**

**After each command, hit the
return key.**



This causes R to execute it.

Try it out...

Type the following at the command prompt:

Simple calculations

> 1 + 1

Pre-defined functions

> abs(-11)

Assign a value to a variable name

> x <- 128.5

Display content of variables

> x

**After each
command, hit
the return key.**



R Key Concepts

- **Variable:** A storage space in memory that has a name and can hold data.
`x <- 12.5` # x is a variable. We use <- to assign a value to it.
- **Function:** pre-written code that performs a specific task and can be executed by "calling" the function.
Write the function's name, followed by parentheses. Inside the parenthesis, pass variables or literal values to the function code (function arguments).
 - `abs(x)`: the absolute value of x
 - `log2(x)`: the base 2-logarithm of x
 - `q()`: to quit R
- **Operator:** a special function for arithmetic, logical or other operations. eg : `+`, `-`, `*`, `/`, `^`, ...
- **Numeric constant:** a number, such as 128.5
- **Character constant:** a text sequence, such a "Hello" (enclosed in quotes)

Working directory - P5S

R can read and write files. The **working directory** is the folder on your computer where it will look for files.

See the current working directory:

```
> getwd()
```

Change the working directory to any **existing folder** on your hard drive or system using `setwd()` and the file path, e.g.

```
> setwd("D:/R_exercises/")
```

R scripts

A script is a file that contains commands to be executed in succession. Write your code into a script and save it

- to have documentation later of what you did
- to be able to re-use the code and create variations
- for easy execution

Writing scripts (.R file)

- Create a new script using **File > New File > R script**.
Don't forget to save your script often.
- By default, scripts are saved into the working directory.
- Files can be saved to other locations (**File -> Save As...**)

Send code from a script to the console

Run **individual lines**, one by one:

- In RStudio: put the cursor anywhere in a line, hit

Ctrl + enter (Windows)

Cmd + return (Mac)

or click the "Run" button

Tip: You can also run a **part of a line** or **multiple lines**: Highlight the code, then proceed as above

Let's practice - 2

1) Get familiar with the different windows of Rstudio

- Console
- Environment / history
- Script
- Files / Help

2) Prepare your first script

- Open a new script file
- Save it as ex1.R
- Paste the following code, and use Ctrl+enter
- (cmd+enter on Mac) to execute each line

#ex1.R

```
# First Steps, ex 1
w <- 3
h <- 0.5
area <- w * h
area
```

R scripts

Tips:

- **Most of your code should be saved in scripts.**
- **You will often want to execute individual lines of code interactively while you are writing it.**
- **You will often want to run the entire script after it is finished and debugged.**

Closing or switching projects

- Close a project:
File -> Close
- Switch to another project, which will close the current one:
File -> Open Project...
- Open another project and keep the current one open as well:
File -> Open Project in New Session...

Reopening an R project from a file

You can access your R project directly from your hard drive

- Find the `.Rproj` file and open it (double click on many systems).
- Rstudio will automatically start if it is not already running.

Workspace (.Rdata) and History (.Rhistory) Options

- When closing or switching projects, the workspace and the history are automatically cleared.

Your RStudio project can be configured to save your workspace and history to a file upon closing a project - or not.

Menu:

Tools -> Global Options -> General (set default for all projects)

Tools -> Project Options -> General (settings for one project)

**CAUTION: .Rdata files can be very large.
Save only when you have space on your hard drive!**

Workspace

The workspace is the internal memory where R stores the objects you created during the session.

Explore your workspace using the command line:

- To list what is in your workspace, type
> ls()
- To remove (delete) all objects from the workspace, type
> rm(list=ls())

Explore your workspace using Rstudio's GUI:

- See the upper right quadrant, tab "Environment": all objects are listed
- To remove all objects from the workspace, click the broom icon.



Saving the Workspace (.RData file)

Before quitting R, you have the option of saving everything that is currently in your workspace to a file.

- To save your entire workspace, use **save.image()** and pass a file path of your choice as argument

```
> save.image("workspace_1.RData")
```

- To save only specific R objects from the workspace, use **save()**, pass all objects you want to save and a file path of your choice

```
> save(x, y, my_sum, file="my_precious_objects.RData")
```

Saving the workspace (.RData file)

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```
> save(x, y, my_sum, file="my_precious_objects.RData")
```

.Rdata files are not human readable.

Loading a saved workspace (.RData file)

After re-starting R, you can load the workspace or the saved objects again.

- Use **load()** with the path to the desired .Rdata file as argument

```
> load("workspace_1.RData")
```

Loading a saved the workspace (.RData file)

After re-starting R, you can load the workspace or the saved objects again.

- Use **load()** with the path to the desired .Rdata file as argument

```
> load("workspace_1.RData")
```

.Rdata files are useful when

- **you want to start at the next session where you left off, without recalculating what you already did**
- **you need to pass R objects to another person**

Find help

- R includes an extensive help system (like **man** in UNIX), which can be accessed directly from the command prompt.
- Use **help()** or **?** to display the help page for a function, operator or other element

```
> ?paste
```

```
> help(paste)
```

```
> ?"^"
```

```
> help("^")
```

- Use **help.search()** or **??** to search for help pages related to a term

```
> help.search("print")
```

```
> ??print
```

Find help

- R includes an extensive help system (like **man** in UNIX), which can be accessed directly from the command prompt.
- Use **help()** or **?** to display the help page for a function, operator or other element

```
> help(paste)
```

```
> ?paste
```

Note: no quotes needed

```
> help("^")
```

```
> ?"^"
```

Note: quotes required!
Single or double quotes will work

- Use **help.search()** or **??** to search for help pages related to a term

```
> help.search("print")
```

```
> ??print
```

Find help

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- Use **help()** or **?** to display the help page for a function, operator or other element

```
> help(paste)
```

```
> ?paste
```

```
> help("^")
```

```
> ?"^"
```

**Being able to use the help
efficiently is arguably
50% of what makes a
programmer**

- Use **help.search()** or **??** to search for help pages related to a term

```
> help.search("print")
```

```
> ??print
```

Tip: Google is your friend !

Packages

- Sets of related functions (and sometimes data sets)
- A small number of packages are part of the basic R installation.
- Many, many packages are developed by the user community and can be installed later as needed.
- There are **two main repositories** that provide R packages of interest in the life sciences. Their content can be browsed on the web :
 - **CRAN** (Comprehensive R Archive Network, <http://cran.r-project.org/>) : the main R repository, with over 16000 packages (October 2020).
 - **Bioconductor** (<http://www.bioconductor.org>) : a separate project specialized in the analysis of high-throughput genomic and transcriptomic data, with over 1900 packages (October 2020).

Installing packages from CRAN

Install packages from CRAN with the `install.packages()` function

```
> install.packages("stringi") # character string manipulations
```

Installation necessary only once until you upgrade R to a new version.

Using functions from packages (I)

Once a package is installed, its content is NOT automatically available for use in your current R session.

`library()` loads the package for the current session.

It is good practice to load all needed packages at the top of a script.

```
# My Script

library(limma)
library(DESeq2)
library(MASS)
library(ggplot2)

# Here my data analysis begins
...
```

Using functions from packages (II)

Alternative: If you need just one function from a package, you can run it without loading the package, using the operator `::`.

```
stringi::stri_length("abc") # use the function stri_length  
                             # from the package stringi  
                             # without having run library(stringi)
```

Session information

- **R.version.string** prints the currently used R version.
- **sessionInfo()** prints version information about R and attached or loaded packages.

Tip: This information is useful when you want to redo an analysis later, generate a report, or post a question on an online forum, ...

Working at the command prompt in RStudio

Shortcuts for both R console and RStudio:

- **TAB key** for command auto completion.
- **Up** and **down arrows** to scroll through the command history.
- **Ctrl-I** to clear console window.

Shortcuts specific to Rstudio:

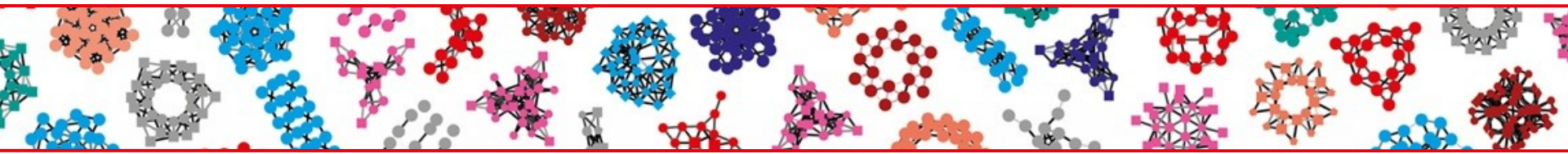
- **Ctrl-1** and **Ctrl-2** to jump between the script and the console windows (shift focus).

Support for incomplete statements (R console and Rstudio):

- If you hit return while your statement is incomplete, (e.g. , with an opening parenthesis but without closing one) the command prompt will change to "+" instead of the usual >. This indicates that R is waiting for you to complete the command.

In a nutshell

- R and RStudio environments
 - [Use of R project](#) to gather all documents related to a project together.
 - The [working directory](#) becomes the [directory of your project](#).
 - Possibility to [save/load workspace](#) (.Rdata file) containing your objects.
 - Possibility to [save/load history of commands](#) (.Rhistory file).
 - [Help](#) and [packages](#) available.
 - More info on how to set up your R environment (if not using R projects) in the extra slides.
- Now let's get familiar with R syntax and objects.



03

Getting started with R syntax and objects

R Basic Data Types

- Variables we have seen so far can hold one value. This value can be of different types. Use `mode()` to display it.

The three most common data types:

- **Numeric:**

- A number stored with decimal point. (Decimal point need not be displayed).

Examples: 0, 5, 55.2, -11.111

- in some contexts this data type is labeled "double"
- integers, stored without decimal points, exists but rarely used.

- **Character:**

A text sequence. Must be enclosed in quotes " ". (Single quotes work, too).

Examples: "1a++", "Hello World", "s", "99"

- **Logical:** TRUE or FALSE (This is binary. No other possible values).

R syntax

Syntax refers to the spelling and "grammar" rules of a programming language.

A few important points :

- **Case sensitive:** R differentiates between small letters and CAPITALS.
- Statements can be separated by a **newline** or by a semicolon ";"
(for better readability, a newline after each statement is almost always preferable)
- Long statements can be written **on multiple lines**
- R has no strict rules about including or omitting **blank spaces** between elements, as long as the code is unambiguous.
- Make your spacing consistent and think about readability.

The **#** character stands for **comments**. Anything after a **#** **on a line is ignored by R**. Write comments into your code to explain what it does.

R objects

An object is a storage space that takes (or contains) a value, a data structure or a section of code.

- All elements of an R statement can be thought of as objects.

Variables are objects containing data.

Functions are objects containing code.

Allowed names for objects

Object names can consist of letters, numbers, dots and underscores.

- Cannot start with a number.
- Cannot contain operators (including hyphen).
- Best to start with letter

Examples:

x

mydata1

mydata.normalized

n_times

The assignment operator "<-" (or equivalent: "=")

- P5S

We can use either the symbol "<-" or "=" to assign values to objects. Stick to one for consistency.

- Create an object:

```
> x <- 10 # Create object x, assign the value 10 to it  
# NB: This does the same as x = 10
```

- Change the value of an existing object:

```
> x <- 25 # x has the value 10; overwrite it
```

- Set one object to equal the value of another object:

```
> myNumber <- 15  
> x <- myNumber # Both x and myNumber now contain 15
```

- Modify the content of an object:

```
> x <- x + sqrt(16) # add the square root of 16 to x
```


Using functions (I)

- Functions are called with parentheses () after the function name
- Arguments are the input to functions, passed inside the ()
 - > ls() # no argument - list objects in workspace
 - > sqrt(81) # one argument - square root of the input
 - > rep(1,5) # two arguments - repeat the number 1 5 times
- Arguments have names (specified in the function definition). Function calls can be made with unnamed or named arguments or a mix of both. Use "=" for named arguments.
 - > rep(x=1, times=5) # Named args. Equivalent to rep(1,5)
 - > rep(1, times=5) # Mixed. Equivalent to rep(1,5)

Check R help (?function_name) to see which arguments are expected by a function.

Using functions (II)

Many functions take more than one argument

- If unnamed, arguments must be listed in correct order (association by position).
- If named, arguments can be passed in arbitrary order (association by name).

```
> write.table(object, "outfile.txt", TRUE)
```

```
> write.table(object, append=TRUE, file="outfile.txt")
```

Unnamed arguments: must appear in their correct position

Named arguments: their position does not matter

Using functions (III)

Some functions have arguments with **default values**.

Example: function **round()**

Usage (from R Help): `round(x, digits = 0)`



**default
value**

Arguments with default values **can be omitted** in the function call; the default value is then used. Arguments without default values **cannot be omitted**.

```
> round(2.011)    #rounding to 0 digits after decimal point  
[1] 2             # (default value)
```

```
> round(2.011, 2) #rounding to 2 digits after decimal point  
[1] 2.01
```

Let's practice – 3

For all exercises, feel free to use

- cheat sheets provided*
- R help (? at command prompt)*

Go back to your script file, then:

- 1) Assign the values 6.7 and 56.3 to variables **a** and **b**, respectively.
- 2) Calculate $(2*a)/b + (a*b)$ and assign the result to variable **x**. Display the content of **x**.
- 3) Find out how to compute the square root of variables. Compute the square roots of **a** and **b** and of the ratio **a/b**.
- 4) a) Calculate the logarithm to the base 2 of **x** (i.e., $\log_2 x$).
b) Calculate the natural logarithm of **x** (i.e., $\log_e x$).

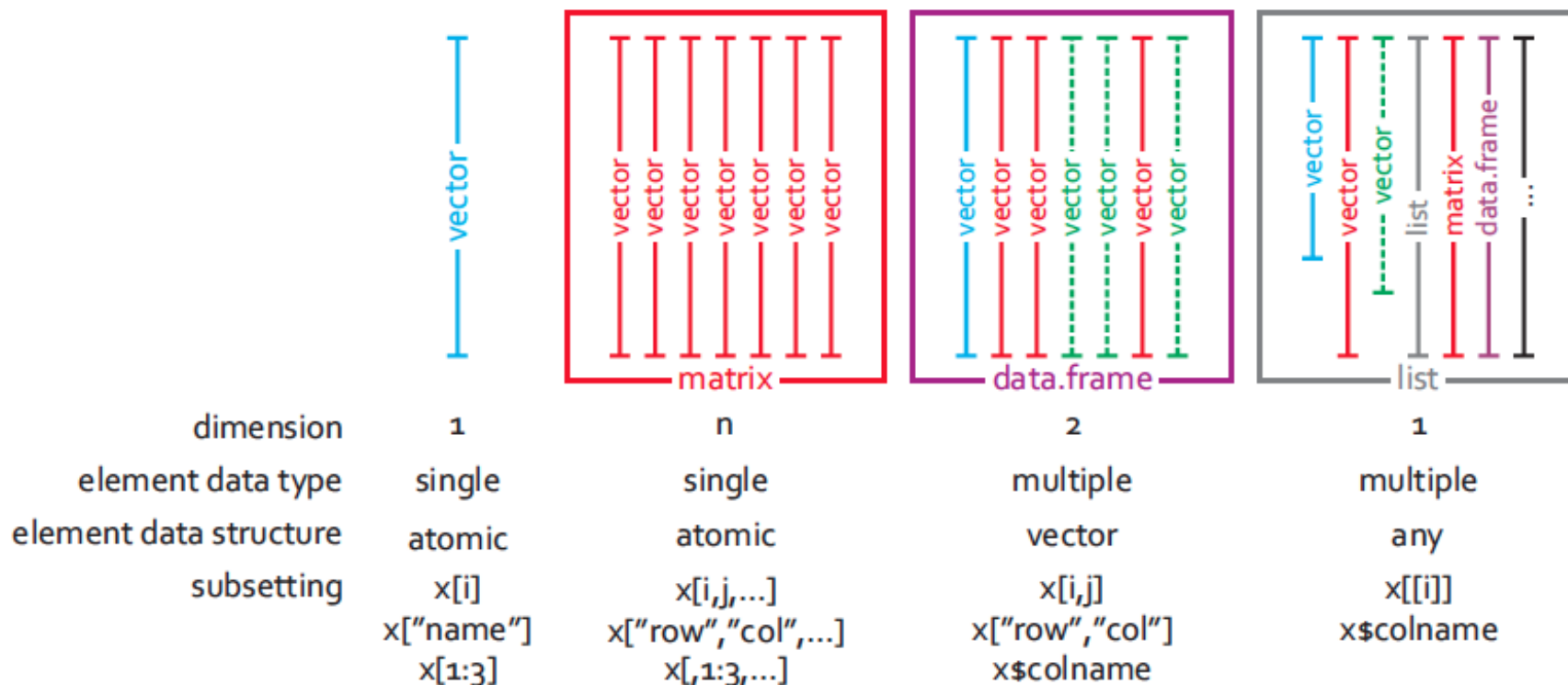
Common object classes

- **vector** – a series of data, all of the same type
- **matrix** – multiple columns of same length, all must have the same type of data
- **data frame** – multiple columns of same length, can be mix of data types
- **list** – a collection of objects; can be of different classes and different sizes
- **function** – a command to perform a specific task

Data objects

Function objects

Graphical view on data object classes



From M. Stadler

Detecting data types and object classes

The function **class()** is useful when we are not sure what kind of object we are dealing with.

- for **vectors**, returns the **basic data type** of its elements ("numeric", "character", "logical", ...)

similar to mode() but slightly more fine-grained

- recognizes "integer" as different from "numeric"
- recognizes factors (categorical variables)

- for all **other objects** covered on previous slide, returns their **class** ("matrix", "data.frame", "list", "function", ...)

Creating objects: Vectors

Vector: A series of data, all of the same type

- Create a vector using `c()`
`height_in_cm <- c(180, 167, 199)`
- Create a vector using `c()` where each element has a name
`height_in_cm <- c(Mia=180, Paul=167, Ed=199)`
- Access elements of a vector using `[]`
`height_in_cm[1]` # get the first element
`height_in_cm[c(1,3)]` # get the 1st and 3rd element
`height_in_cm["Paul"]` # get the element named "Paul"

Scalars in R (the simple variables we have seen so far) can be thought of as vectors of length 1.

Creating objects: More ways to generate vectors

- **:** (colon operator)

```
> a <- 1:10
```

```
> a
```

```
[1] 1 2 3 4 5 6 7 8 9 10
```

- **seq()** for sequences with any step size

```
> s <- seq(4,10,2) #start at 4, end at 10, step by 2
```

```
> s
```

```
[1] 4 6 8 10
```

-

Vector manipulation

Applying operators to vector result in element-wise operations

```
> a
```

```
[1] 1 2 3 4
```

```
> a * 2 # multiply by 2 each element of a
```

```
[1] 2 4 6 8
```

```
> a + c(12,10,12,10) # addition of the elements in 2 vectors
```

```
[1] 13 12 15 14
```

Many functions take a vector as argument and either perform an element-wise operation or return a single value

```
> log2(a) # compute the logarithm in base 2 of each elements
```

```
[1] 0.000000 1.000000 1.584963 2.000000
```

```
> mean(a) # compute mean of the elements
```

```
[1] 2.5
```

Coercion

- P5S

- All elements of a vector must be the same type
- If combining different types, they will be coerced to the most flexible type
 - least to most flexible are: logical < numeric < character

Example:

```
> vec <- c(12, "twelve", TRUE) #combine 3 data types
> vec                          #all are coerced to character
```

```
[1] "12" "twelve" "TRUE"
```

```
> class(vec)
```

```
[1] "character"
```

Coercion

- We can coerce an existing vector to another type using the functions **as.logical()**, **as.numeric()**, **as.character()**.
- Example: Coerce a logical vector to numeric
Values are converted to 1 (for TRUE) and 0 (for FALSE)
 - we can use **as.numeric()** for explicit coercion
 - we can use mathematical functions on logical vectors, coercion to numeric happens automatically

```
> x <- c(FALSE, FALSE, TRUE)
```

```
> as.numeric(x)
```

```
> sum(x)    # number that are true
```

```
> mean(x)   # proportion that are true
```

Factors

- A **factor** is a vector containing values from a limited set; used for storing categorical data.
- Example: Genotype of mouse individuals
`> genotype <- factor(c("WT", "WT", "Mut2", "Mut1", "Mut2"))`

The available values in a factor are called **levels**.
Extract them:

```
> levels(genotype)
[1] "Mut1" "Mut2" "WT"
```

- Convert the factor back to a character vector:
`> geno <- as.character(genotype)`

Factors with custom sorted levels

- By default, factor levels are sorted alphabetically.
- We can specify a different sorting.

- Example: Genotype of mouse individuals,

```
> genotype <- factor(c("WT", "WT", "Mut2", "Mut1", "Mut2"),  
  levels = c("WT", "Mut1", "Mut2"))
```

```
> levels(genotype)
```

```
[1] "WT" "Mut1" "Mut2"
```

Levels are sorted the way we wanted

Let's practice - 4

- 5) Create two vectors, **vector_a** and **vector_b**, containing the values from -5 to 5 and from 10 down to 0 , respectively.
- 6) Calculate the (element-wise) sum, difference and product between the elements of **vector_a** and **vector_b**.
- 7) a) Calculate the sum of elements in **vector_a**.
b) Calculate the overall sum of elements in both **vector_a** and **vector_b**.
- 8) Identify the smallest and the largest value among both **vector_a** and **vector_b**.
- 9) Compute the overall mean of the values among both **vector_a** and **vector_b**.

Hint: Each task in exercises 6-9 can be performed in a single statement per vector

Operators (most commonly used ones)

- **Arithmetic**

+, -, *, /, ^

- **Comparison**

>, <, <=, >=, == (equal to), != (not equal to)

- **Logical**

! (negation), & (AND), | (OR)

- **Other**

%in% (in operator)

Comparisons, logical operators and %in%
always return logical values!
(TRUE, FALSE)

Operators returning logical values: examples

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

```
[1] FALSE FALSE FALSE FALSE TRUE TRUE FALSE
```

```
> !c(1,3,5,1,2,2,1) < 2
```

```
[1] FALSE TRUE TRUE FALSE TRUE TRUE FALSE
```

```
> table(!c(1,3,5,1,2,2,1) < 2)
```

```
#FALSE TRUE
```

```
#3      4
```

```
> c("Fred", "Marc", "Dan", "Ali") %in%  
  c("Dan", "Geoff", "Ali")
```

```
[1] FALSE FALSE TRUE TRUE
```

Missing Values

- R distinguishes between
 - **NA** (not available)
 - **NaN** (not a number) and
 - **Inf** (Infinity, result of division by 0)
- Use the functions **is.na()** and **is.nan()** to detect them.

Missing Values: Examples (I)

Missing values are usually represented by NA:

```
> y <- c(1,2,3,4,5,NA,NA)
```

NA's interfere with many functions:

```
> mean(y)
```

```
[1] NA
```

Arguments often exist to remove NA's before calculation

```
> mean(y, na.rm=TRUE)
```

```
[1] 3
```

Alternatively, use **omit.na()** to remove NAs from the data

```
> mean(omit.na(y))
```

```
[1] 3
```

Missing Values: Examples (II)

```
> x <- c(1:3, NA, 0/0, 1/0) ; x # a vector to play with  
[1] 1 2 3 NA NaN Inf
```

```
> is.na(x) #detects NAs and NaNs from x  
[1] FALSE FALSE FALSE TRUE TRUE FALSE
```

```
> is.nan(x) # detects only NaNs from x  
[1] FALSE FALSE FALSE FALSE TRUE FALSE
```

```
> x > 2 # what if we try to compare NA and NaN to a number?  
[1] FALSE FALSE TRUE NA NA TRUE
```

```
> x[!is.na(x)] # removes NAs and NaNs from x  
[1] 1 2 3 Inf
```

Creating objects: Data Frames

- P5S

data frame: multiple columns of same length, can be mix of data types

```
>name <- c("Joyce", "Chaucer", "Homer")  
>status <- c("dead", "deader", "deadeast")  
>reader_rating <- c(55, 22, 100)
```

Create a data frame using the function **data.frame()**

```
>df <- data.frame(name, status, reader_rating)  
>df
```

	name	status	reader_rating
1	Joyce	dead	55
2	Chaucer	deader	22
3	Homer	deadeast	100

Creating objects: Lists

List: a collection of objects; can be of different classes and different sizes

Create a few objects:

```
> v <- c(0.4, 0.9, 0.6)
> m <- cbind(c(1,1), c(2,1))
> d <- data.frame(name = c("Ed", "Lisa"), age = c(61, 71),
  stringsAsFactors = FALSE)
```

Unnamed list - collect these objects in a list, using the function **list()**:

```
> l <- list(v, m, d)
```

Named list - collect these objects in a list with named elements:

```
> l_with_names <- list(myvector=v, mymatrix=m, mydata=d)
```

Accessing Data Elements

Data frame:

```
>df[2, 2]    # gets the element on row 2 in column 2
>df[, 2]      # gets all elements in column 2
>df[1:3, ]    # gets rows 1,2,3
>df[, c(1,3)] # gets columns 1 and 3
>df[, c("name", "reader_rating")] # gets columns "name"
                                   # and "reader_rating"
>df$name      # gets column "name"
```

list:

```
>l[[1]]      # gets the first object
>l_with_names[["myvec"]] # gets the object named "myvec"
>l_with_names$myvec    # gets the object named "myvec", too
```

Accessing Names of Data Elements

matrix and data frame:

```
>rownames(df)      # gets the row names
```

```
>colnames(df)      # gets the column names
```

```
>rownames(df) <- c("J", "C", "H") # sets/overwrites row names
```

list:

```
>names(l_with_names) # gets the list elements' names
```

```
>names(l_with_names) <- c("A", "B", "C") # overwrites names
```

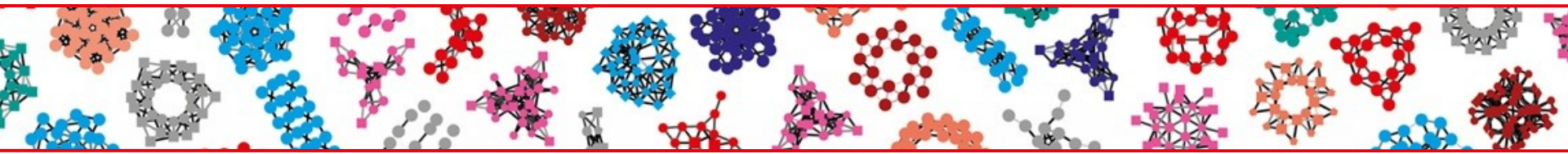

Let's practice – 5

- 1) Install and load the package “MASS”
- 2) The following command line loads the **bacteria** data.frame present in the MASS package. Execute it:
- 3) `> data(bacteria)`
- 4) What are the names of the columns of the **bacteria** data.frame ?
- 5) Use the `[]` , to select in **bacteria** rows 100 to 119 in the “trt” column.

Optional : 5) use comparison operators to count how many rows correspond to a “placebo” treatment (“trt” column).

In a nutshell

- Everything in R is an object.
- Using R is all about creating and manipulating data objects using functions (which are themselves objects).
 - Objects can be assigned to a name
 - Objects have a class (data frame, matrix, list etc)
 - Data values inside objects have different data storage modes (numeric, character, logical)
- We covered many ways to generate data (create objects).
- Now, let's import some data !



04 Formatting your data

Example of a dataset

2467	RB_2	BE-04		1	1	1	1	1	12	0	55	M	1
2468	HB_2	BE-05		1	1	1	1	1	13	1	66	M	1
2482	WO_2	ZH-01		1	1	1	1	1	7	1	64	M	1
2484	HW_2	ZH-04		1	1	1	1	1	5	1	50	M	1
2485	BD_2	ZH-05		1	1	1	1	1	6	0	53	F	1
2486	BH_2	ZH-06		1	1	1	1	1	9	1	48	F	1
2487	AW_2	ZH-07		1	1	1	1	1	9	0	53	M	1
2488	AJN_2	ZH-08		1	1	1	1	1	5	0	35	M	1
2489	KO_2	ZH-09		1	0	1	1	1	54	0	59	M	1
2490	BS_2	ZH-11		1	0	1	1	1	150	0	59	M	1
2491	KPR_3	ZH-12		1	1	1	1	1	5	0	32	M	1
2492	CB_3	ZH-13		1	0	1	1	0	6	0	37	F	1
2493	RM_3	ZH-14		1	0	1	1	1	63	0	39	M	1
2496	BR_2	ZH-17		1	1	1	1	1	5	0	61	F	1
2497	SP_2_0	2497		1		0	0			1	58	M	1
2498	NA_2_0	2498		1		0	0			0	54	M	1
2499	GK_2_0	2499		1		0	0			1	68	M	1
2500	HiB_2_0	2500		1		0	0			1	62	M	1
2501	BI_2	2501		1		0	0			0	70	F	1
2502	WJ_2	2502		1		0	0			1	59	M	1
2503	BP_3	2503	autopsy	1		0	0			0	61	M	1
2504	UA_2_0	2504		1		0	0			0	35	F	1
2505	GE_1	2505		0		0	0			1	65	F	1
2506	TS_2	2506		1		0	0			0	50	M	1
2507	HV_2_0	2507		1		0	0			0	65	F	1
2508	TI_3	2508		1		0	0			1	31	F	1
2509	TI_4_0	2509	Rec 2508	0		0	0			1	31	F	1
2510	GE_2_0	2510	Rec 2505	1		0	0			1	67	F	0
2511	SI_2	ZH-18		1	1	1	1	1	5	0	24	F	1
2512	BH_3	ZH-06.1	Rec 2486	0		1	0			1	50	F	1
2513	CG_2	2513		1		0	0			0	63	M	1
1152	NCHI152	NCHI152	Xenograft			0				1		hXenograft	1
1154	NCHI154	NCHI154	Xenograft			0				1		hXenograft	1
1155	NCHI155	NCHI155	Xenograft			0				1		hXenograft	1
1157	NCHI157	NCHI157	Xenograft	1		1			5	1		hXenograft	1

Difficult to use with any statistical program

Courtesy of Frédéric Schütz

Prepare your data outside of R

Before using R and importing the dataset you collected from an experiment, you need to know **how to format** it properly, so R can read it.

A spreadsheet program such as Excel or OpenOffice can be used for data entry and simple manipulation.

Three precepts of tidy data:

1. Each variable forms a column.
2. Each observation forms a row.
3. Each type of observational unit forms a table.

<http://www.ucd.ie/ecomodel/pdf/TidyData.pdf>

Example of well-formatted dataset

	A	B	C	D
1	chr	pos	minor	major
2	1	123369	A	C
3	1	138369	G	T
4	1	153369	T	C
5	1	168369	C	T
6	1	183369	G	A
7	1	198369	T	A
8	5	228369	G	A
9	5	258369	G	A
10	5	288369	A	G
11	5	318369	C	A
12	5	348369	A	T

- A header line with variable names
- 4 variables, one in each column
- One observation per row, here SNP info

Example of well-formatted dataset

	A	B	C	D
1	chr	pos	minor	major
2	1	123369	A	C
3	1	138369	G	T
4	1	153369		
5	1	168369		
6	1	183369		
7	1	198369		
8	5	228369		
9	5	258369		
10	5	288369		
11	5	318369		
12	5	348369		

Glossary for the snp data

snp single nucleotide polymorphism

chr chromosome

pos position

minor minor allele (a minority of individuals have this letter at this position)

major major allele (a majority of individuals have this letter at this position)

Formatting recommendations – checklist

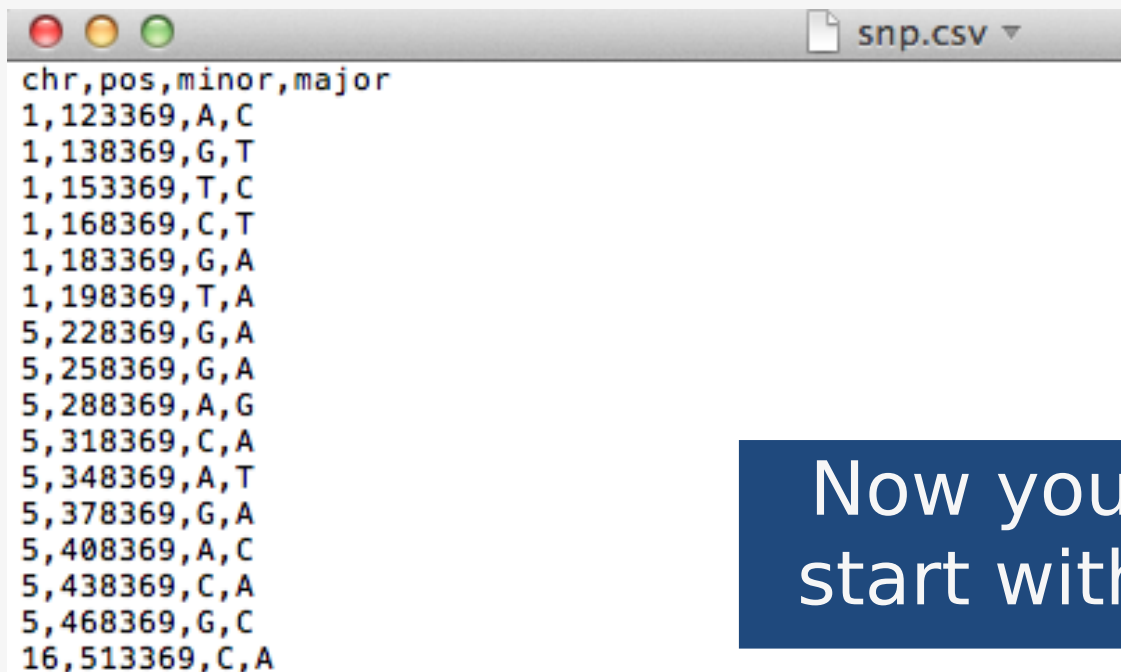
- If you work with spreadsheets, the first row is usually reserved for the header.
- The first column may or may not be an ID column.
- Remove blank spaces from column names and in fields. If you want to concatenate words, insert a "." or "_" between words.
- Avoid column names containing symbols other than "." and "_".
- Short names are preferred over longer names.
- Delete any comments or other content in the spreadsheet that are not part of the data table but are above, below or beside the data table.
- Make sure that any missing values in your data set are indicated with NA. (Check spelling! N.A. or n.a. does not work.)

Other recommendations

- If you're using a spreadsheet, keep a copy of the **original data** as it was provided to you. Prepare a new, "cleaned" version for your data analysis.
- Do not include columns that you do not need for your analysis.
- Have data backups!

Saving your data

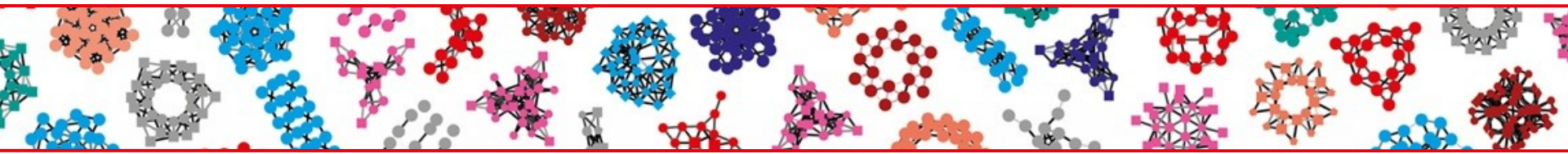
- **Export** the spreadsheet to your computer in a text file format:
 - csv (comma separated values) format, with file extension **.CSV** OR
 - tsv (tab separated values) format, with file extension **.txt** or **.tsv**



The screenshot shows a text editor window with the title bar 'snp.csv'. The content is a list of SNPs in CSV format, with columns for chromosome (chr), position (pos), minor allele, and major allele. The data is as follows:

chr	pos	minor	major
1	123369	A	C
1	138369	G	T
1	153369	T	C
1	168369	C	T
1	183369	G	A
1	198369	T	A
5	228369	G	A
5	258369	G	A
5	288369	A	G
5	318369	C	A
5	348369	A	T
5	378369	G	A
5	408369	A	C
5	438369	C	A
5	468369	G	C
16	513369	C	A

Now you are ready to
start with the analysis



05 Importing/exporting data into R

Importing Data

- Most widely used R base function for data import: **read.table()**
 - **reads** a formatted text file
 - **imports it as a data frame**
 - **many** options, to accommodate most text files (e.g., csv, tsv).
- To read an entire data frame from a file, it should have:
 1. **a header line** containing the names of all variables
 - (not obligatory but preferable)
 2. **one line per row**, with values for each variable
 - (missing values should be indicated using **NA**)
 3. Items must be **separated by the same separator symbol**
 - (most common: **, ; \t**)

Importing Data – the 2 questions

- Where is the file I want to import?
 - Look for your file in the file system.
 - Note its path: the succession of folders to access it
- Where is my session being run (working directory)?
 - use **getwd()** (**recommendation: should be the project directory**)

File Paths in R

File paths can be specified as a string with `'/'` as separator:

```
"C:/Users/Leo/courses/data/snp.csv"
```

Or with a little help from the function `file.path()`:

```
file.path("C:", "Users", "Leo", "courses", "data", "snp.csv")
```

WARNINGS:

On Windows : replace `'\'` by `'/'`,

Escape spaces in file names : `' ' → '\ '`

File Paths in R - relative

R understands "." and ".." for relative file paths

- . is the current directory (=working directory)

- .. is the parent directory

`"/data/snp.csv"` # file "snps.csv" in subfolder "data" of current directory

`"/../snp.csv"` # file "snps.csv", 2 levels up from current directory

Also works with `file.path()`:

`file.path("../", "../", "snp.csv")`

Note: `"data/snp.csv"` is equivalent to `"/data/snp.csv"` for most purposes

Importing Data – File Paths

`read.table()` needs to know where the file is located.

- **Data file is in the working directory:** file name suffices.

`"snp.csv"`

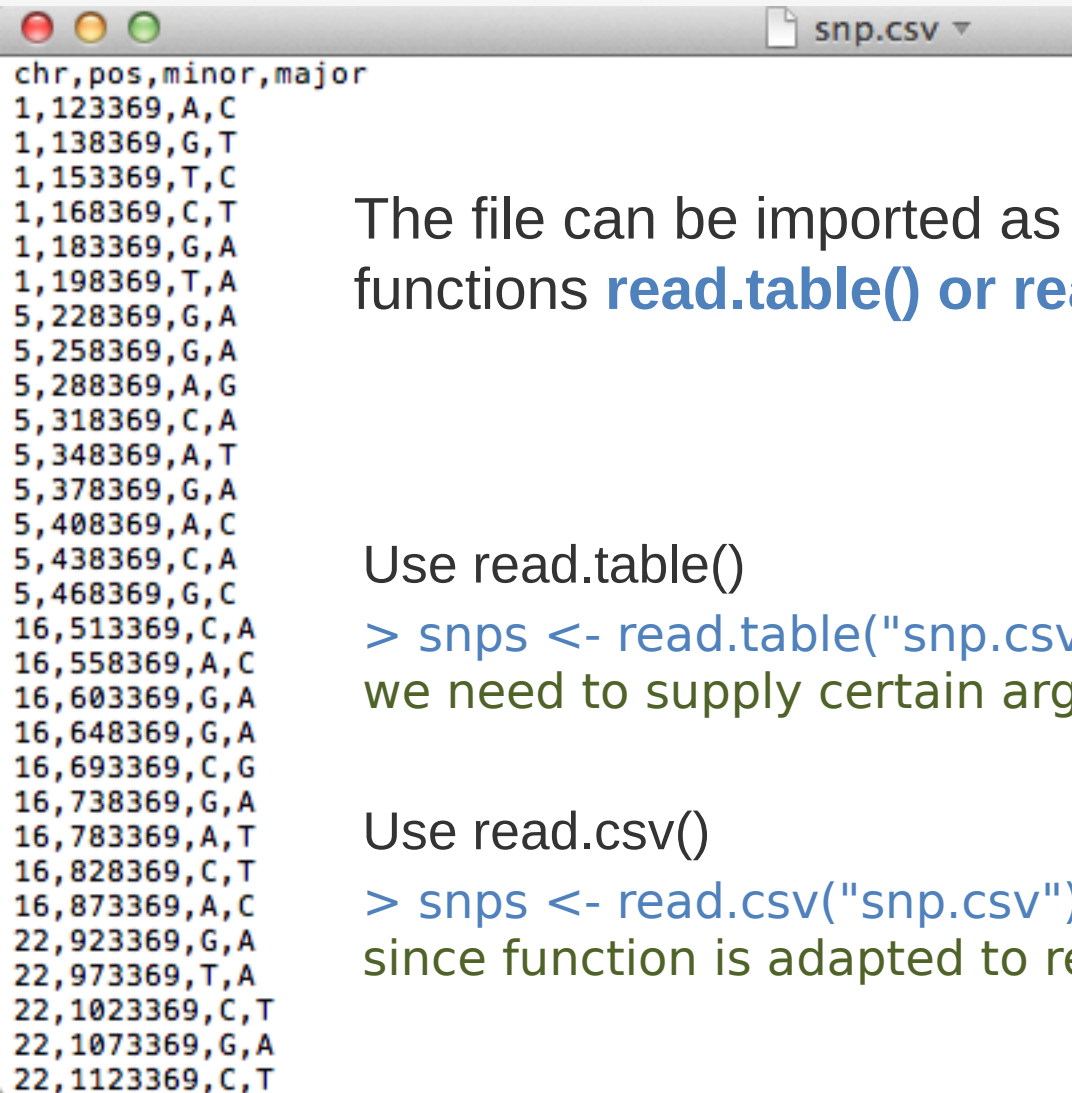
- **Data file is in a sub-folder of working directory:** It's easy to use a **relative path**. (Great option for **projects** shared with **others**).

`data/snp.csv` or

`file.path("data", "snp.csv")`

- **Data file is somewhere else or you are not working inside a project:** it's safest to use an **absolute path** (but can be more painful to specify!).

`"C:/Users/Leo/courses/data/snp.csv"`



```
chr,pos,minor,major
1,123369,A,C
1,138369,G,T
1,153369,T,C
1,168369,C,T
1,183369,G,A
1,198369,T,A
5,228369,G,A
5,258369,G,A
5,288369,A,G
5,318369,C,A
5,348369,A,T
5,378369,G,A
5,408369,A,C
5,438369,C,A
5,468369,G,C
16,513369,C,A
16,558369,A,C
16,603369,G,A
16,648369,G,A
16,693369,C,G
16,738369,G,A
16,783369,A,T
16,828369,C,T
16,873369,A,C
22,923369,G,A
22,973369,T,A
22,1023369,C,T
22,1073369,G,A
22,1123369,C,T
```

The file can be imported as a data frame using the functions **read.table()** or **read.csv()**

Use read.table()

```
> snps <- read.table("snp.csv", header=TRUE, sep=",") #  
we need to supply certain arguments
```

Use read.csv()

```
> snps <- read.csv("snp.csv") # arguments can be omitted  
since function is adapted to reading .csv
```

ON PRACTICE:

Both are valid, use the one you can remember

Importing Data

- Important optional arguments of **read.table()**, **read.csv()**, **read.delim()** (check ?read.table for more):
 - **header** : TRUE/FALSE → whether the file contains **column names** on the first line. **Default in read.table()** is FALSE.
 - **Default in read.csv()** and **read.delim()** is TRUE.
- **sep** allows to specify **the field separator** character (e.g. "," or tab "\t").
 - Default in read.table()** is any white space characters (i.e. space, tab, newline and carriage return).
 - Default in read.csv()** is comma. **Default in read.delim()** is tab.
- **colClasses** : manually setting each variable data type

ON PRACTICE:

When in doubt, use **help(read.table)**

It is very important to check that **data you asked R to import** is **the data you wanted**.

> head(snps) # shows first 6 rows (tail(snps) - shows last 6 rows)

	chr	pos	minor	major
1	1	123369	A	C
2	1	138369	G	T
3	1	153369	T	C
4	1	168369	C	T
5	1	183369	G	A
6	1	198369	T	A

> dim(snps)

[1] 40 4

> nrow(snps);ncol(snps)

[1] 40

[1] 4

```
> colnames(snps) # column names
```

```
[1] "chr" "pos" "minor" "major"
```

```
> str(snps) # structure of the data frame
```

```
'data.frame': 40 obs. of 4 variables:
```

```
$ chr : int 1 1 1 1 1 1 5 5 5 5 ...
```

```
$ pos : int 123369 138369 153369 168369 183369 198369  
228369 258369 288369 318369 ...
```

```
$ minor: chr "A" "G" "T" "C" ...
```

```
$ major: chr "C" "T" "C" "T" ...
```

R made its best guess for data types.

- Are they what we need?
- Do we wish to convert any variables to factors?

Setting factor variables

Convert categorical variables to factors as needed.

```
> snps$chr <- factor(snps$chr, levels=c("1","5","16","22"))  
> snps$minor <- factor(snps$minor)  
> snps$major <- factor(snps$major)
```

```
> str(snps) # structure of the data frame  
'data.frame': 40 obs. of 4 variables:  
 $ chr : Factor w/ 4 levels "1","5","16","22": 1 1 1 1 1 1 2 2 2 2 ...  
 $ pos : int 123369 138369 153369 168369 183369 198369 228369  
258369 288369 318369 ...  
 $ minor: Factor w/ 4 levels "A","C","G","T": 1 3 4 2 3 4 3 3 1 2 ...  
 $ major: Factor w/ 4 levels "A","C","G","T": 2 4 2 4 1 1 1 1 3 1 ...
```

```
> summary(snps)
```

chr	pos	minor	major
1 : 6	Min. : 123369	A: 9	A:17
5 : 9	1st Qu. : 340869	C:10	C:10
16: 9	Median : 715869	G:15	G: 4
22:16	Mean : 777869	T: 6	T: 9
	3rd Qu. : 1185869		
	Max. : 1673369		

Customizing summaries of data

`tapply()` generates custom summaries of your data using :

- X: a column you want to aggregate (of any data type)
- INDEX: a factor column, or list of factor columns, to structure the aggregation
- FUN: a function to be applied to X (mean, sd, min, max, length, median, range, quantiles...), separately for each grouping indicated by INDEX

```
> tapply(X=snps$pos, INDEX=snps$chr, FUN=min)
```

```
1      5      16      22
123369 228369 513369 923369
```

In each chromosome, find the smallest (min) position number where a SNP is located.

Accessing parts of the data

```
> snps[2,] # 2nd row
  chr  pos minor major
2  1 138369   A     T
```

```
> snps[, "minor"] # column named "minor"
[1] A A T C G T G G A C A G A C G C A G G C G A C A G T C G C C T A G G
T G T C G A
Levels: A C G T
```

```
> snps[1:3, c(1,3)] # 3 first rows, 1st and 3rd column
  chr minor
1  1     A
2  1     G
3  1     T
```



```
> snps$chr # vector of chromosomes, equivalent to snps[, 1]
[1] 1 1 1 1 1 1 5 5 5 5 5 5 5 5 5 16 16 16 16 16 16 16 16 16
22 22 22
[28] 22 22 22 22 22 22 22 22 22 22 22 22 22 22 22
```

```
> snps$chr[40] # chromosome of the last row
[1] 22
```

Accessing parts of the data

- `subset()` is a powerful function which allows you to **subset your data** by **specific columns and values** in those columns. **Logical operators** can be used within the subset.

```
> subset(snps, chr==1) # keeps only the snps where chr is 1
```

	chr	pos	minor	major
1	1	123369	A	C
2	1	138369	G	T
3	1	153369	T	C
4	1	168369	C	T
5	1	183369	G	A
6	1	198369	T	A

```
> subset(snps, chr==1 & major=="A") # keeps only the snps in  
chr 1 with an "A" as major allele
```

	chr	pos	minor	major
5	1	183369	G	A
6	1	198369	T	A

```
> subset(snps, chr==1 & major=="A" & minor=="T")  
# keeps only the snps in chr 1 with an "A" as major allele and T  
as minor allele
```

	chr	pos	minor	major
6	1	198369	T	A

Data reshaping : adding rows and columns -P5S

- Rows and columns of data can be **added** using the functions **rbind()** and **cbind()**, respectively.

- Add a row to the SNP data:

```
> snps_updated <- rbind(snps,  
  data.frame(chr=22, pos=1723369, minor="A", major="T"))
```

- Add a column of indexes to the SNP data:

```
> idx <- 1:nrow(snps)  
> snps_mod <- cbind(idx,snps)
```

Always check afterwards that your new dataset is what you expect, the same way you did after you imported the original one

Data reshaping : removing a column

- Remove the new column of indexes, using exclusion (-) or column extraction

```
> snps_orig <- snps_mod[,-1] # remove the first column  
> head(snps_orig) # check resulting data
```

or

```
> snps_orig <- snps_mod[,2:dim(snps_mod)[2]] # extract all columns  
that you want to keep (from the 2nd to the last)  
  
> head(snps_orig) # check resulting data
```

Exporting data to a file

The functions **write.table()** and **write.csv()** allow to write a data frame to a file.

Example:

```
> write.table(snps_updated, file="snps_updated.csv", quote=FALSE,  
sep="," ,row.names=FALSE)
```

- Important optional arguments (check ?write.table for more):
 - **File** : output file path and name .
 - **append** : **append to an existing file** (default is FALSE).
 - **quote** : whether the elements of **character or factor columns** should be surrounded by double quotes (default is TRUE).
 - **sep** : **field separator** to be used, e.g., comma (",") or tab ("\t").
 - **row.names** : whether or not the row names are written (default is TRUE). Alternatively, accepts a character vector with **new row names to be written**.
 - **col.names** : specifies whether **the column names** are written (default is TRUE)

In a nutshell

- How to **import** data into data frames (R's typical container for data)
- How to **check** the imported data, **summarize** it , **access** part of it, and **manipulate** it.
- How to **export** data to files
- Next step tomorrow: How to represent data graphically?

Let's practice - 6

A dataset from mouse experiments at 18 weeks is available in the file ***mice_data_mod.csv*** (courtesy of F Schutz and F. Preitner). Let's explore the dataset to see what it contains.

- 1) Open a new script file in R studio, and save it.
- 2) Have look at the csv file in R studio's file explorer. What do you need to check in order to be able to read in the file correctly?
- 3) Read the file into R, assign its content to object "mice_data". Examine the object.
- 4) How many observations and variables does the dataset have?
- 5) What is the structure of the dataset? What are the names and classes of the variables?
- 6) Which variables appear to be categorical? Convert them to factors.
- 7) Get the summary statistics of "mice_data"

Let's practice – 6bis

- 8) Use the function `table()` to compute the number of observations in different mouse groups. a) How many mice are included of each genotype (WT, KO)? b) How many mice are included per diet (HFD, CHOW)? c) Make a 2x2 table by genotype and diet crossed.
- 9) Compute the means and standard deviations for WT and KO mouse weights using `tapply()`. Then do the same for CHOW and HFD groups.
- 10) Isolate the observations for the mice on high fat diet (HFD) using `subset()`. Compute a summary statistics just for the weights of the subset. Then do the same for the mice on regular chow diet (CHOW). Export the data of each subgroup to a csv file.
- 11) Look at the results from the two previous exercises. What does this initial exploration of the data suggest about mouse weights?