
The R Bioc Book

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Preface

Who is this book for?

- People who want to learn data science
- People who want to teach data science
- People who want to learn how to teach data science
- People who want to learn how to learn data science

Why this book?

This book is a collection of resources for learning R and Bioconductor. It is meant to be largely self-directed, but for those looking to teach data science, it can also be used as a guide for structuring a course. Material is a bit variable in terms of difficulty, prerequisites, and format which is a reflection of the organic creation of the material.

Students are encouraged to work with others to learn the material. Instructors are encouraged to use the material to create a course that is tailored to the needs of their students and to spend lots of time in 1:1 and small groups to support students in their learning. See below for additional thoughts on adult learning and how it relates to this material.

Adult learners

Adult Learning Theory, also known as Andragogy, is the concept and practice of designing, developing, and delivering instructional experiences for adult learners. It is based on the belief that adults learn differently than children, and thus, require

Adult learners

distinct approaches to engage, motivate, and retain information (Center 2016). The term was first introduced by Malcolm Knowles, an American educator who is known for his work in adult education (Knowles, Holton, and Swanson 2005).

One of the fundamental principles of Adult Learning Theory is that adults are self-directed learners. This means that we prefer to take control of our own learning process and set personal goals for themselves. We are motivated by our desire to solve problems or gain knowledge to improve our lives (see Figure 1). As a result, educational content for adults should be relevant and applicable to real-life situations. Furthermore, adult learners should be given opportunities to actively engage in the learning process by making choices, setting goals, and evaluating their progress.

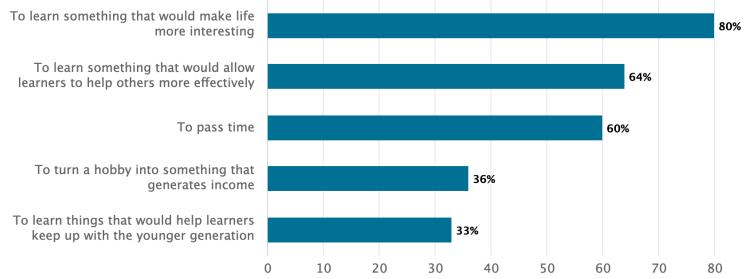


Figure 1.: Why do adults choose to learn something?

Another key aspect of Adult Learning Theory is the role of experience. We bring a wealth of experience to the learning process, which serves as a resource for new learning. We often have well-established beliefs, values, and mental models that can influence our willingness to accept new ideas and concepts. Therefore, it is essential to acknowledge and respect our shared and unique past experiences and create an environment where we all feel comfortable sharing our perspectives.

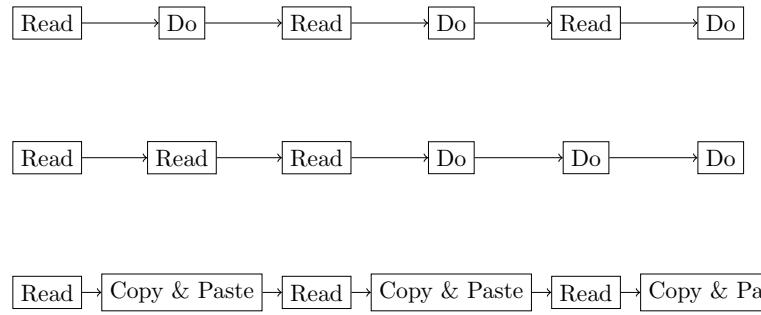
To effectively learn as a group of adult learners, it is crucial to establish a collaborative learning environment that promotes open communication and fosters trust among participants. We all appreciate and strive for a respectful and supportive atmosphere where we can express our opinions without fear of judgment. Instructors should help facilitate discussions, encourage peer-to-peer interactions, and incorporate group activities and

Adult learners

collaboration to capitalize on the collective knowledge of participants.

Additionally, adult learners often have multiple responsibilities outside of the learning environment, such as work and family commitments. As a result, we require flexible learning opportunities that accommodate busy schedules. Offering a variety of instructional formats, such as online modules, self-paced learning, or evening classes, can help ensure that adult learners have access to education despite any time constraints.

Adult learners benefit from a learner-centered approach that focuses on the individual needs, preferences, and interests of each participant can greatly enhance the overall learning experience. In addition, we tend to be more intrinsically motivated to learn when we have a sense of autonomy and can practice and experiment (see Figure 2) with new concepts in a safe environment.



Understanding Adult Learning Theory and its principles can significantly enhance the effectiveness of teaching and learning as adults. By respecting our autonomy, acknowledging our experiences, creating a supportive learning environment, offering flexible learning opportunities, and utilizing diverse teaching methods, we can better cater to the unique needs and preferences of adult learners.

In practice, that means that we will not be prescriptive in our approach to teaching data science. We will not tell you what to do, but rather we will provide you with a variety of options and you can choose what works best for you. We will also provide you with a variety of resources and you can choose where to focus your time. Given that we cannot possibly cover

Figure 2.: How to stay stuck in data science (or anything). The “Read-Do” loop tends to deliver the best results. Too much reading between doing can be somewhat effective. Reading and simply copy-paste is probably the least effective. When working through material, experiment. Try to break things. Incorporate your own experience or applications whenever possible.

Adult learners

everything, we will provide you with a framework for learning and you can fill in the gaps as you see fit. A key component of our success as adult learners is to gain the confidence to ask questions and problem-solve on our own.

Part I.

Introduction

1. Introducing R and RStudio

Questions

- What is R?
- Why use R?
- Why not use R?
- Why use RStudio and how does it differ from R?

Learning Objectives

- Know advantages of analyzing data in R
- Know advantages of using RStudio
- Be able to start RStudio on your computer
- Identify the panels of the RStudio interface
- Be able to customize the RStudio layout

1.1. Introduction

In this chapter, we will discuss the basics of R and RStudio, two essential tools in genomics data analysis. We will cover the advantages of using R and RStudio, how to set up RStudio, and the different panels of the RStudio interface.

1.2. What is R?

R([https://en.wikipedia.org/wiki/R_\(programming_language\)](https://en.wikipedia.org/wiki/R_(programming_language))) is a programming language and software environment designed for statistical computing and graphics. It is widely used by statisticians, data scientists, and researchers for data analysis

Learning Objectives

and visualization. R is an open-source language, which means it is free to use, modify, and distribute. Over the years, R has become particularly popular in the fields of genomics and bioinformatics, owing to its extensive libraries and powerful data manipulation capabilities.

The R language is a dialect of the S language, which was developed in the 1970s at Bell Laboratories. The first version of R was written by Robert Gentleman and Ross Ihaka and released in 1995 (see [this slide deck](#) for Ross Ihaka's take on R's history). Since then, R has been continuously developed by the R Core Team, a group of statisticians and computer scientists. The R Core Team releases a new version of R every year.

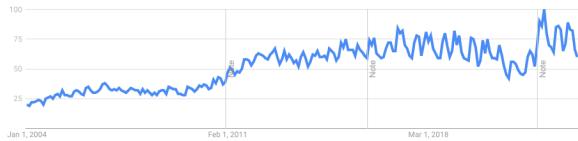


Figure 1.1.: Google trends showing the popularity of R over time based on Google searches

1.3. Why use R?

There are several reasons why R is a popular choice for data analysis, particularly in genomics and bioinformatics. These include:

1. **Open-source:** R is free to use and has a large community of developers who contribute to its growth and development. [What is “open-source”?](#)
2. **Extensive libraries:** There are thousands of R packages available for a wide range of tasks, including specialized packages for genomics and bioinformatics. These libraries have been extensively tested and are available for free.
3. **Data manipulation:** R has powerful data manipulation capabilities, making it easy (or at least possible) to clean, process, and analyze large datasets.
4. **Graphics and visualization:** R has excellent tools for creating high-quality graphics and visualizations that can be customized to meet the specific needs of your analysis.

Learning Objectives

In most cases, graphics produced by R are publication-quality.

5. **Reproducible research:** R enables you to create reproducible research by recording your analysis in a script, which can be easily shared and executed by others. In addition, R does not have a meaningful graphical user interface (GUI), which renders analysis in R much more reproducible than tools that rely on GUI interactions.
6. **Cross-platform:** R runs on Windows, Mac, and Linux (as well as more obscure systems).
7. **Interoperability with other languages:** R can interact with FORTRAN, C, and many other languages.
8. **Scalability:** R is useful for small and large projects.

I can develop code for analysis on my Mac laptop. I can then install the *same* code on our 20k core cluster and run it in parallel on 100 samples, monitor the process, and then update a database (for example) with R when complete.

1.4. Why not use R?

- R cannot do everything.
- R is not always the “best” tool for the job.
- R will *not* hold your hand. Often, it will *slap* your hand instead.
- The documentation can be opaque (but there is documentation).
- R can drive you crazy (on a good day) or age you prematurely (on a bad one).
- Finding the right package to do the job you want to do can be challenging; worse, some contributed packages are unreliable.]{}]
- R does not have a meaningfully useful graphical user interface (GUI).

Learning Objectives

1.5. R License and the Open Source Ideal

R is free (yes, totally free!) and distributed under GNU license. In particular, this license allows one to:

- Download the source code
- Modify the source code to your heart's content
- Distribute the modified source code and even charge money for it, but you must distribute the modified source code under the original GNU license]{}{}

This license means that R will always be available, will always be open source, and can grow organically without constraint.

1.6. RStudio

RStudio is an integrated development environment (IDE) for R. It provides a graphical user interface (GUI) for R, making it easier to write and execute R code. RStudio also provides several other useful features, including a built-in console, syntax-highlighting editor, and tools for plotting, history, debugging, workspace management, and workspace viewing. RStudio is available in both free and commercial editions; the commercial edition provides some additional features, including support for multiple sessions and enhanced debugging

1.6.1. Getting started with RStudio

To get started with RStudio, you first need to install both R and RStudio on your computer. Follow these steps:

1. Download and install R from the [official R website](#).
2. Download and install RStudio from the [official RStudio website](#).
3. Launch RStudio. You should see the RStudio interface with four panels.

Learning Objectives

1.6.2. The RStudio Interface

RStudio's interface consists of four panels (see Figure 1.2):

- **Console** This panel displays the R console, where you can enter and execute R commands directly. The console also shows the output of your code, error messages, and other information.
- **Source** This panel is where you write and edit your R scripts. You can create new scripts, open existing ones, and run your code from this panel.
- **Environment** This panel displays your current workspace, including all variables, data objects, and functions that you have created or loaded in your R session.
- **Plots, Packages, Help, and Viewer** These panels display plots, installed packages, help files, and web content, respectively.

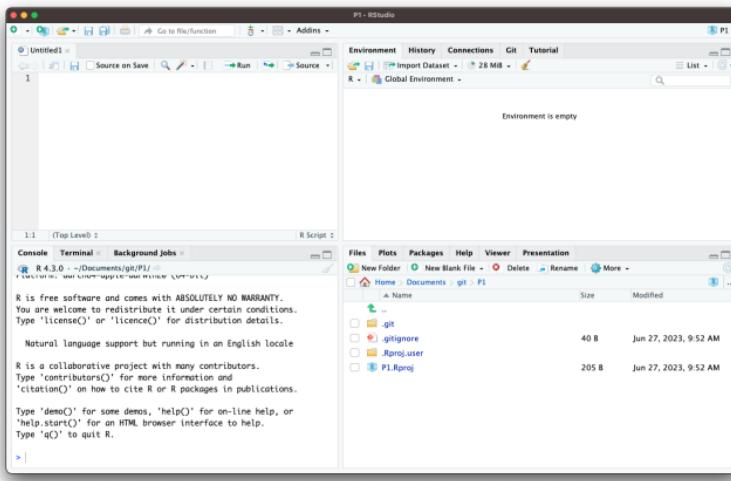


Figure 1.2.: The RStudio interface. In this layout, the **source** pane is in the upper left, the **console** is in the lower left, the **environment** panel is in the top right and the **viewer/help/files** panel is in the bottom right.

i Do I need to use RStudio?

No. You can use R without RStudio. However, RStudio makes it easier to write and execute R code, and it provides several useful features that are not available in the

Learning Objectives

basic R console. Note that the only part of RStudio that is actually interacting with R directly is the console. The other panels are simply providing a GUI that enhances the user experience.

💡 Customizing the RStudio Interface

You can customize the layout of RStudio to suit your preferences. To do so, go to **Tools > Global Options > Appearance**. Here, you can change the theme, font size, and panel layout. You can also resize the panels as needed to gain screen real estate (see Figure 1.3).

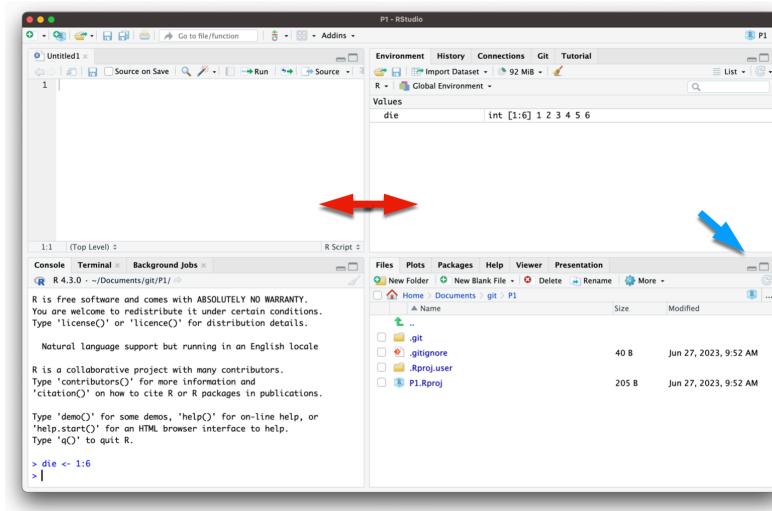


Figure 1.3.: Dealing with limited screen real estate can be a challenge, particularly when you want to open another window to, for example, view a web page. You can resize the panes by sliding the center divider (red arrows) or by clicking on the minimize/maximize buttons (see blue arrow).

In summary, R and RStudio are powerful tools for genomics data analysis. By understanding the advantages of using R and RStudio and familiarizing yourself with the RStudio interface, you can efficiently analyze and visualize your data. In the following chapters, we will delve deeper into the functionality of R, Bioconductor, and various statistical methods to help you gain a comprehensive understanding of genomics data analysis.

2. R mechanics

2.1. Learning objectives

- Be able to start R and RStudio
- Learn to interact with the R console
- Know the difference between expressions and assignment
- Recognize valid and invalid R names
- Know how to access the R help system
- Know how to assign values to variables, find what is in R memory, and remove values from R memory

2.2. Starting R

How to start R depends a bit on the operating system (Mac, Windows, Linux) and interface. In this course, we will largely be using an Integrated Development Environment (IDE) called *RStudio*, but there is nothing to prohibit using R at the command line or in some other interface (and there are a few).

2.3. *RStudio*: A Quick Tour

The RStudio interface has multiple panes. All of these panes are simply for convenience except the “Console” panel, typically in the lower left corner (by default). The console pane contains the running R interface. If you choose to run R outside RStudio, the interaction will be *identical* to working in the console pane. This is useful to keep in mind as some environments, such as a computer cluster, encourage using R without RStudio.

- Panes
- Options

2. R mechanics

- Help
- Environment, History, and Files

2.4. Interacting with R

The only meaningful way of interacting with R is by typing into the R console. At the most basic level, anything that we type at the command line will fall into one of two categories:

1. Assignments

```
x = 1  
y <- 2
```

2. Expressions

```
1 + pi + sin(42)
```

```
[1] 3.225071
```

The assignment type is obvious because either the The <- or = are used. Note that when we type expressions, R will return a result. In this case, the result of R evaluating `1 + pi + sin(42)` is 3.2250711.

The standard R prompt is a “>” sign. When present, R is waiting for the next expression or assignment. If a line is not a complete R command, R will continue the next line with a “+”. For example, typing the following with a “Return” after the second “+” will result in R giving back a “+” on the next line, a prompt to keep typing.

```
1 + pi +  
sin(3.7)
```

```
[1] 3.611757
```

R can be used as a glorified calculator by using R expressions. Mathematical operations include:

- Addition: +

2. R mechanics

- Subtraction: -
- Multiplication: *
- Division: /
- Exponentiation: ^
- Modulo: %%

The `^` operator raises the number to its left to the power of the number to its right: for example 3^2 is 9. The modulo returns the remainder of the division of the number to the left by the number on its right, for example 5 modulo 3 or $5 \% 3$ is 2.

2.4.1. Expressions

```
5 + 2  
28 %% 3  
3^2  
5 + 4 * 4 + 4 ^ 4 / 10
```

Note that R follows order-of-operations and groupings based on parentheses.

```
5 + 4 / 9  
(5 + 4) / 9
```

2.4.2. Assignment

While using R as a calculator is interesting, to do useful and interesting things, we need to assign *values* to *objects*. To create objects, we need to give it a name followed by the assignment operator `<-` (or, entirely equivalently, `=`) and the value we want to give it:

```
weight_kg <- 55
```

`<-` is the assignment operator. Assigns values on the right to objects on the left, it is like an arrow that points from the value to the object. Using an `=` is equivalent (in nearly all cases). Learn to use `<-` as it is good programming practice.

2. R mechanics

Objects can be given any name such as `x`, `current_temperature`, or `subject_id` (see below). You want your object names to be explicit and not too long. They cannot start with a number (`2x` is not valid but `x2` is). R is case sensitive (e.g., `weight_kg` is different from `Weight_kg`). There are some names that cannot be used because they represent the names of fundamental functions in R (e.g., `if`, `else`, `for`, see [here](#) for a complete list). In general, even if it's allowed, it's best to not use other function names, which we'll get into shortly (e.g., `c`, `T`, `mean`, `data`, `df`, `weights`). When in doubt, check the help to see if the name is already in use. It's also best to avoid dots (.) within a variable name as in `my.dataset`. It is also recommended to use nouns for variable names, and verbs for function names.

When assigning a value to an object, R does not print anything. You can force to print the value by typing the name:

```
weight_kg
```

```
[1] 55
```

Now that R has `weight_kg` in memory, which R refers to as the “global environment”, we can do arithmetic with it. For instance, we may want to convert this weight in pounds (weight in pounds is 2.2 times the weight in kg).

```
2.2 * weight_kg
```

```
[1] 121
```

We can also change a variable's value by assigning it a new one:

```
weight_kg <- 57.5  
2.2 * weight_kg
```

```
[1] 126.5
```

2. R mechanics

This means that assigning a value to one variable does not change the values of other variables. For example, let's store the animal's weight in pounds in a variable.

```
weight_lb <- 2.2 * weight_kg
```

and then change `weight_kg` to 100.

```
weight_kg <- 100
```

What do you think is the current content of the object `weight_lb`, 126.5 or 220?

You can see what objects (variables) are stored by viewing the Environment tab in Rstudio. You can also use the `ls()` function. You can remove objects (variables) with the `rm()` function. You can do this one at a time or remove several objects at once. You can also use the little broom button in your environment pane to remove everything from your environment.

```
ls()  
rm(weight_lb, weight_kg)  
ls()
```

What happens when you type the following, now?

```
weight_lb # oops! you should get an error because weight_lb no longer exists!
```

2.5. Rules for Names in R

R allows users to assign names to objects such as variables, functions, and even dimensions of data. However, these names must follow a few rules.

- Names may contain any combination of letters, numbers, underscore, and “.”
- Names may not start with numbers, underscore.
- R names are case-sensitive.

2. R mechanics

Examples of valid R names include:

```
pi  
x  
camelCaps  
my_stuff  
MY_Stuff  
this.is.the.name.of.the.man  
ABC123  
abc1234asdf  
.hi
```

2.6. Resources for Getting Help

There is extensive built-in help and documentation within R. A separate page contains a collection of [additional resources](#).

If the name of the function or object on which help is sought is known, the following approaches with the name of the function or object will be helpful. For a concrete example, examine the help for the `print` method.

```
help(print)  
help('print')  
?print
```

If the name of the function or object on which help is sought is *not* known, the following from within R will be helpful.

```
help.search('microarray')  
RSiteSearch('microarray')  
apropos('histogram')
```

There are also tons of online resources that Google will include in searches if online searching feels more appropriate.

I strongly recommend using `help("newfunction")` for all functions that are new or unfamiliar to you.

There are also many open and free resources and reference guides for R.

2. R mechanics

- [Quick-R](#): a quick online reference for data input, basic statistics and plots
- R reference card [PDF](#) by Tom Short
- Rstudio [cheatsheets](#)

3. Up and Running with R

In this chapter, we’re going to get an introduction to the R language, so we can dive right into programming. We’re going to create a pair of virtual dice that can generate random numbers. No need to worry if you’re new to programming. We’ll return to many of the concepts here in more detail later.

To simulate a pair of dice, we need to break down each die into its essential features. A die can only show one of six numbers: 1, 2, 3, 4, 5, and 6. We can capture the die’s essential characteristics by saving these numbers as a group of values in the computer. Let’s save these numbers first and then figure out a way to “roll” our virtual die.

3.1. The R User Interface

The RStudio interface is simple. You type R code into the bottom line of the RStudio console pane and then click Enter to run it. The code you type is called a *command*, because it will command your computer to do something for you. The line you type it into is called the *command line*.

When you type a command at the prompt and hit Enter, your computer executes the command and shows you the results. Then RStudio displays a fresh prompt for your next command. For example, if you type `1 + 1` and hit Enter, RStudio will display:

```
> 1 + 1  
[1] 2  
>
```

3. Up and Running with R

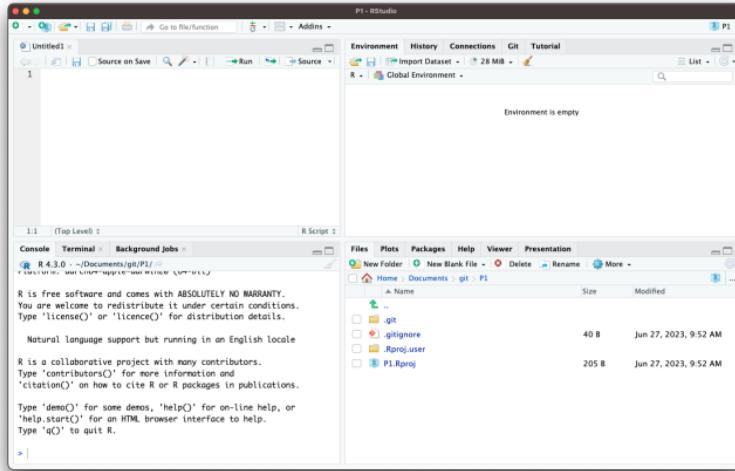


Figure 3.1.: Your computer does your bidding when you type R commands at the prompt in the bottom line of the console pane. Don't forget to hit the Enter key. When you first open RStudio, the console appears in the pane on your left, but you can change this with **File > Tools > Global Options** in the menu bar.

You'll notice that a [1] appears next to your result. R is just letting you know that this line begins with the first value in your result. Some commands return more than one value, and their results may fill up multiple lines. For example, the command 100:130 returns 31 values; it creates a sequence of integers from 100 to 130. Notice that new bracketed numbers appear at the start of the second and third lines of output. These numbers just mean that the second line begins with the 14th value in the result, and the third line begins with the 25th value. You can mostly ignore the numbers that appear in brackets:

```
> 100:130
[1] 100 101 102 103 104 105 106 107 108 109 110 111 112
[14] 113 114 115 116 117 118 119 120 121 122 123 124 125
[25] 126 127 128 129 130
```

Tip

The colon operator (:) returns every integer between two integers. It is an easy way to create a sequence of numbers.

3. Up and Running with R

When do we compile?

In some languages, like C, Java, and FORTRAN, you have to compile your human-readable code into machine-readable code (often 1s and 0s) before you can run it. If you've programmed in such a language before, you may wonder whether you have to compile your R code before you can use it. The answer is no. R is a dynamic programming language, which means R automatically interprets your code as you run it.

If you type an incomplete command and press Enter, R will display a + prompt, which means R is waiting for you to type the rest of your command. Either finish the command or hit Escape to start over:

```
> 5 -  
+  
+ 1  
[1] 4
```

If you type a command that R doesn't recognize, R will return an error message. If you ever see an error message, don't panic. R is just telling you that your computer couldn't understand or do what you asked it to do. You can then try a different command at the next prompt:

```
> 3 % 5  
Error: unexpected input in "3 % 5"  
>
```

Tip

Whenever you get an error message in R, consider googling the error message. You'll often find that someone else has had the same problem and has posted a solution online. Simply cutting-and-pasting the error message into a search engine will often work

3. Up and Running with R

Once you get the hang of the command line, you can easily do anything in R that you would do with a calculator. For example, you could do some basic arithmetic:

```
2 * 3
```

```
[1] 6
```

```
4 - 1
```

```
[1] 3
```

```
# this obeys order-of-operations  
6 / (4 - 1)
```

```
[1] 2
```

💡 Tip

R treats the hashtag character, `#`, in a special way; R will not run anything that follows a hashtag on a line. This makes hashtags very useful for adding comments and annotations to your code. Humans will be able to read the comments, but your computer will pass over them. The hashtag is known as the *commenting symbol* in R.

❗ Cancelling commands

Some R commands may take a long time to run. You can cancel a command once it has begun by pressing `ctrl + c` or by clicking the “stop sign” if it is available in Rstudio. Note that it may also take R a long time to cancel the command.

3. Up and Running with R

3.1.1. An exercise

That's the basic interface for executing R code in RStudio. Think you have it? If so, try doing these simple tasks. If you execute everything correctly, you should end up with the same number that you started with:

1. Choose any number and add 2 to it.
2. Multiply the result by 3.
3. Subtract 6 from the answer.
4. Divide what you get by 3.

```
10 + 2
```

```
[1] 12
```

```
12 * 3
```

```
[1] 36
```

```
36 - 6
```

```
[1] 30
```

```
30 / 3
```

```
[1] 10
```

3.2. Objects

Now that you know how to use R, let's use it to make a virtual die. The `:` operator from a couple of pages ago gives you a nice way to create a group of numbers from one to six. The `:` operator returns its results as a **vector** (we are going to work with vectors in more detail), a one-dimensional set of numbers:

3. Up and Running with R

```
1:6  
## 1 2 3 4 5 6
```

That's all there is to how a virtual die looks! But you are not done yet. Running `1:6` generated a vector of numbers for you to see, but it didn't save that vector anywhere for later use. If we want to use those numbers again, we'll have to ask your computer to save them somewhere. You can do that by creating an R *object*.

R lets you save data by storing it inside an R object. What is an object? Just a name that you can use to call up stored data. For example, you can save data into an object like `a` or `b`. Wherever R encounters the object, it will replace it with the data saved inside, like so:

```
a <- 1  
a
```

```
[1] 1
```

```
a + 2
```

```
[1] 3
```

i What just happened?

1. To create an R object, choose a name and then use the less-than symbol, `<`, followed by a minus sign, `-`, to save data into it. This combination looks like an arrow, `<-`. R will make an object, give it your name, and store in it whatever follows the arrow. So `a <- 1` stores 1 in an object named `a`.
2. When you ask R what's in `a`, R tells you on the next line.
3. You can use your object in new R commands, too. Since `a` previously stored the value of 1, you're now adding 1 to 2.

3. Up and Running with R

! Assignment vs expressions

Everything that you type into the R console can be assigned to one of two categories:

- Assignments
- Expressions

An expression is a command that tells R to do something. For example, `1 + 2` is an expression that tells R to add 1 and 2. When you type an expression into the R console, R will evaluate the expression and return the result. For example, if you type `1 + 2` into the R console, R will return 3. Expressions can have “side effects” but they don’t explicitly result in anything being added to R memory.

```
5 + 2
```

```
[1] 7
```

```
28 %% 3
```

```
[1] 1
```

```
3^2
```

```
[1] 9
```

```
5 + 4 * 4 + 4 ^ 4 / 10
```

```
[1] 46.6
```

While using R as a calculator is interesting, to do useful and interesting things, we need to assign values to objects. To create objects, we need to give it a name followed by the assignment operator `<-` (or, entirely equivalently, `=`) and the value we want to give it:

```
weight_kg <- 55
```

3. Up and Running with R

So, for another example, the following code would create an object named `die` that contains the numbers one through six. To see what is stored in an object, just type the object's name by itself:

```
die <- 1:6  
die
```

```
[1] 1 2 3 4 5 6
```

When you create an object, the object will appear in the environment pane of RStudio, as shown in Figure 3.2. This pane will show you all of the objects you've created since opening RStudio.

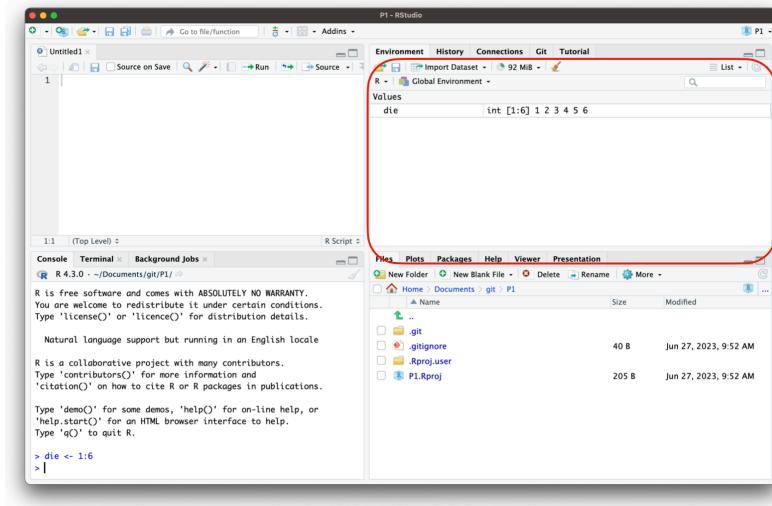


Figure 3.2.: Assignment creates an object in the environment pane.

You can name an object in R almost anything you want, but there are a few rules. First, a name cannot start with a number. Second, a name cannot use some special symbols, like `^`, `!`, `$`, `@`, `+`, `-`, `/`, or `*`:

Good names	Names that cause errors
a	1trial
b	\$
FOO	^mean
my_var	2nd

3. Up and Running with R

Good names	Names that cause errors
.day	!bad

⚠ Capitalization matters

R is case-sensitive, so `name` and `Name` will refer to different objects:

```
> Name = 0
> Name + 1
[1] 1
> name + 1
Error: object 'name' not found
```

The error above is a common one!

Finally, R will overwrite any previous information stored in an object without asking you for permission. So, it is a good idea to *not* use names that are already taken:

```
my_number <- 1
my_number
```

```
[1] 1
```

```
my_number <- 999
my_number
```

```
[1] 999
```

You can see which object names you have already used with the function `ls`:

```
ls()
```

Your environment will contain different names than mine, because you have probably created different objects.

3. Up and Running with R

You can also see which names you have used by examining RStudio's environment pane.

We now have a virtual die that is stored in the computer's memory and which has a name that we can use to refer to it. You can access it whenever you like by typing the word `die`.

So what can you do with this die? Quite a lot. R will replace an object with its contents whenever the object's name appears in a command. So, for example, you can do all sorts of math with the die. Math isn't so helpful for rolling dice, but manipulating sets of numbers will be your stock and trade as a data scientist. So let's take a look at how to do that:

```
die - 1
```

```
[1] 0 1 2 3 4 5
```

```
die / 2
```

```
[1] 0.5 1.0 1.5 2.0 2.5 3.0
```

```
die * die
```

```
[1] 1 4 9 16 25 36
```

R uses *element-wise execution* when working with a *vector* like `die`. When you manipulate a set of numbers, R will apply the same operation to each element in the set. So for example, when you run `die - 1`, R subtracts one from each element of `die`.

When you use two or more vectors in an operation, R will line up the vectors and perform a sequence of individual operations. For example, when you run `die * die`, R lines up the two `die` vectors and then multiplies the first element of vector 1 by the first element of vector 2. R then multiplies the second element of vector 1 by the second element of vector 2, and so on, until every element has been multiplied. The result will be a new vector the same length as the first two {Figure 3.3}.

3. Up and Running with R

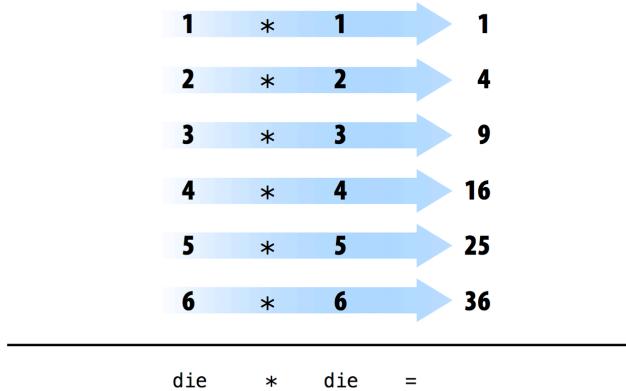


Figure 3.3.: “When R performs element-wise execution, it matches up vectors and then manipulates each pair of elements independently.”

If you give R two vectors of unequal lengths, R will repeat the shorter vector until it is as long as the longer vector, and then do the math, as shown in Figure 3.4. This isn’t a permanent change—the shorter vector will be its original size after R does the math. If the length of the short vector does not divide evenly into the length of the long vector, R will return a warning message. This behavior is known as *vector recycling*, and it helps R do element-wise operations:

```
1:2
```

```
[1] 1 2
```

```
1:4
```

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

```
die
```

```
[1] 1 2 3 4 5 6
```

```
die + 1:2
```

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

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```
die + 1:4
```

Warning in die + 1:4: longer object length is not a multiple of shorter object length

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

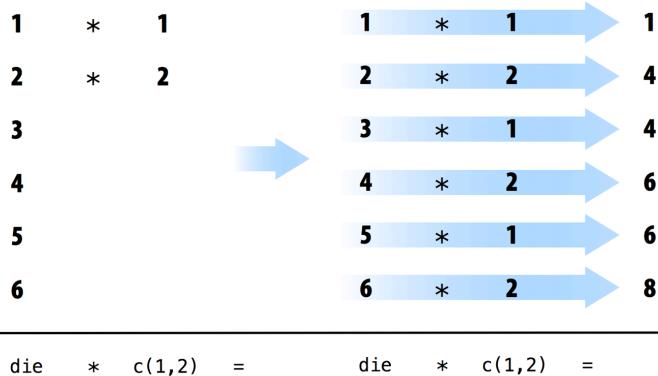


Figure 3.4.: “R will repeat a short vector to do element-wise operations with two vectors of uneven lengths.”

Element-wise operations are a very useful feature in R because they manipulate groups of values in an orderly way. When you start working with data sets, element-wise operations will ensure that values from one observation or case are only paired with values from the same observation or case. Element-wise operations also make it easier to write your own programs and functions in R.

! Element-wise operations are not matrix operations

It is important to know that operations with vectors are not the same that you might expect if you are expecting R to perform “matrix” operations. R can do inner multiplication with the `%*%` operator and outer multiplication with the `%o%` operator:

```
# Inner product (1*1 + 2*2 + 3*3 + 4*4 + 5*5 + 6*6)
die %*% die
# Outer product
die %o% die
```

3. Up and Running with R

Now that you can do math with your `die` object, let's look at how you could "roll" it. Rolling your die will require something more sophisticated than basic arithmetic; you'll need to randomly select one of the die's values. And for that, you will need a *function*.

3.3. Functions

R has many functions and puts them all at our disposal. We can use functions to do simple and sophisticated tasks. For example, we can round a number with the `round` function, or calculate its factorial with the `factorial` function. Using a function is pretty simple. Just write the name of the function and then the data you want the function to operate on in parentheses:

```
round(3.1415)
```

```
[1] 3
```

```
factorial(3)
```

```
[1] 6
```

The data that you pass into the function is called the function's *argument*. The argument can be raw data, an R object, or even the results of another R function. In this last case, R will work from the innermost function to the outermost Figure 3.5.

```
mean(1:6)
```

```
[1] 3.5
```

```
mean(die)
```

```
[1] 3.5
```

3. Up and Running with R

```
round(mean(die))
```

```
[1] 4
```

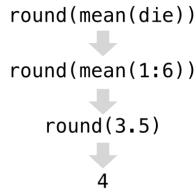


Figure 3.5.: “When you link functions together, R will resolve them from the innermost operation to the outermost. Here R first looks up die, then calculates the mean of one through six, then rounds the mean.”

Returning to our die, we can use the `sample` function to randomly select one of the die’s values; in other words, the `sample` function can simulate rolling the `die`.

The `sample` function takes *two* arguments: a vector named `x` and a number named `size`. `sample` will return `size` elements from the vector:

```
sample(x = 1:4, size = 2)
```

```
[1] 3 4
```

To roll your die and get a number back, set `x` to `die` and sample one element from it. You’ll get a new (maybe different) number each time you roll it:

```
sample(x = die, size = 1)
```

```
[1] 5
```

```
sample(x = die, size = 1)
```

```
[1] 5
```

3. Up and Running with R

```
sample(x = die, size = 1)
```

```
[1] 3
```

Many R functions take multiple arguments that help them do their job. You can give a function as many arguments as you like as long as you separate each argument with a comma.

You may have noticed that I set `die` and `1` equal to the names of the arguments in `sample`, `x` and `size`. Every argument in every R function has a name. You can specify which data should be assigned to which argument by setting a name equal to data, as in the preceding code. This becomes important as you begin to pass multiple arguments to the same function; names help you avoid passing the wrong data to the wrong argument. However, using names is optional. You will notice that R users do not often use the name of the first argument in a function. So you might see the previous code written as:

```
sample(die, size = 1)
```

```
[1] 4
```

Often, the name of the first argument is not very descriptive, and it is usually obvious what the first piece of data refers to anyways.

But how do you know which argument names to use? If you try to use a name that a function does not expect, you will likely get an error:

```
round(3.1415, corners = 2)
## Error in round(3.1415, corners = 2) : unused argument(s) (corners = 2)
```

If you're not sure which names to use with a function, you can look up the function's arguments with `args`. To do this, place the name of the function in the parentheses behind `args`. For example, you can see that the `round` function takes two arguments, one named `x` and one named `digits`:

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```
args(round)
```

```
function (x, digits = 0, ...)  
NULL
```

Did you notice that `args` shows that the `digits` argument of `round` is already set to 0? Frequently, an R function will take optional arguments like `digits`. These arguments are considered optional because they come with a default value. You can pass a new value to an optional argument if you want, and R will use the default value if you do not. For example, `round` will round your number to 0 digits past the decimal point by default. To override the default, supply your own value for `digits`:

```
round(3.1415)
```

```
[1] 3
```

```
round(3.1415, digits = 2)
```

```
[1] 3.14
```

```
# pi happens to be a built-in value in R  
pi
```

```
[1] 3.141593
```

```
round(pi)
```

```
[1] 3
```

You should write out the names of each argument after the first one or two when you call a function with multiple arguments. Why? First, this will help you and others understand your code. It is usually obvious which argument your first input refers to

3. Up and Running with R

(and sometimes the second input as well). However, you'd need a large memory to remember the third and fourth arguments of every R function. Second, and more importantly, writing out argument names prevents errors.

If you do not write out the names of your arguments, R will match your values to the arguments in your function by order. For example, in the following code, the first value, `die`, will be matched to the first argument of `sample`, which is named `x`. The next value, `1`, will be matched to the next argument, `size`:

```
sample(die, 1)
```

```
[1] 3
```

As you provide more arguments, it becomes more likely that your order and R's order may not align. As a result, values may get passed to the wrong argument. Argument names prevent this. R will always match a value to its argument name, no matter where it appears in the order of arguments:

```
sample(size = 1, x = die)
```

```
[1] 3
```

3.3.1. Sample with Replacement

If you set `size = 2`, you can *almost* simulate a pair of dice. Before we run that code, think for a minute why that might be the case. `sample` will return two numbers, one for each die:

```
sample(die, size = 2)
```

```
[1] 1 5
```

3. Up and Running with R

I said this “almost” works because this method does something funny. If you use it many times, you’ll notice that the second die never has the same value as the first die, which means you’ll never roll something like a pair of threes or snake eyes. What is going on?

By default, `sample` builds a sample *without replacement*. To see what this means, imagine that `sample` places all of the values of `die` in a jar or urn. Then imagine that `sample` reaches into the jar and pulls out values one by one to build its sample. Once a value has been drawn from the jar, `sample` sets it aside. The value doesn’t go back into the jar, so it cannot be drawn again. So if `sample` selects a six on its first draw, it will not be able to select a six on the second draw; six is no longer in the jar to be selected. Although `sample` creates its sample electronically, it follows this seemingly physical behavior.

One side effect of this behavior is that each draw depends on the draws that come before it. In the real world, however, when you roll a pair of dice, each die is independent of the other. If the first die comes up six, it does not prevent the second die from coming up six. In fact, it doesn’t influence the second die in any way whatsoever. You can recreate this behavior in `sample` by adding the argument `replace = TRUE`:

```
sample(die, size = 2, replace = TRUE)
```

```
[1] 3 4
```

The argument `replace = TRUE` causes `sample` to sample *with replacement*. Our jar example provides a good way to understand the difference between sampling with replacement and without. When `sample` uses replacement, it draws a value from the jar and records the value. Then it puts the value back into the jar. In other words, `sample` replaces each value after each draw. As a result, `sample` may select the same value on the second draw. Each value has a chance of being selected each time. It is as if every draw were the first draw.

Sampling with replacement is an easy way to create *independent random samples*. Each value in your sample will be a sample

3. Up and Running with R

of size one that is independent of the other values. This is the correct way to simulate a pair of dice:

```
sample(die, size = 2, replace = TRUE)
```

```
[1] 4 2
```

Congratulate yourself; you've just run your first simulation in R! You now have a method for simulating the result of rolling a pair of dice. If you want to add up the dice, you can feed your result straight into the `sum` function:

```
dice <- sample(die, size = 2, replace = TRUE)  
dice
```

```
[1] 4 5
```

```
sum(dice)
```

```
[1] 9
```

What would happen if you call `dice` multiple times? Would R generate a new pair of dice values each time? Let's give it a try:

```
dice
```

```
[1] 4 5
```

```
dice
```

```
[1] 4 5
```

```
dice
```

```
[1] 4 5
```

3. Up and Running with R

The name `dice` refers to a *vector* of two numbers. Calling more than once does not change the favlue. Each time you call `dice`, R will show you the result of that one time you called `sample` and saved the output to `dice`. R won't rerun `sample(die, 2, replace = TRUE)` to create a new roll of the dice. Once you save a set of results to an R object, those results do not change.

However, it *would* be convenient to have an object that can re-roll the dice whenever you call it. You can make such an object by writing your own R function.

3.4. Writing Your Own Functions

To recap, you already have working R code that simulates rolling a pair of dice:

```
die <- 1:6
dice <- sample(die, size = 2, replace = TRUE)
sum(dice)
```

```
[1] 9
```

You can retype this code into the console anytime you want to re-roll your dice. However, this is an awkward way to work with the code. It would be easier to use your code if you wrapped it into its own function, which is exactly what we'll do now. We're going to write a function named `roll` that you can use to roll your virtual dice. When you're finished, the function will work like this: each time you call `roll()`, R will return the sum of rolling two dice:

```
roll()
## 8

roll()
## 3

roll()
## 7
```

3. Up and Running with R

Functions may seem mysterious or fancy, but they are *just another type of R object*. Instead of containing data, they contain code. This code is stored in a special format that makes it easy to reuse the code in new situations. You can write your own functions by recreating this format.

3.4.1. The Function Constructor

Every function in R has three basic parts: a name, a body of code, and a set of arguments. To make your own function, you need to replicate these parts and store them in an R object, which you can do with the `function` function. To do this, call `function()` and follow it with a pair of braces, {}:

```
my_function <- function() {}
```

This function, as written, doesn't do anything (yet). However, it is a valid function. You can call it by typing its name followed by an open and closed parenthesis:

```
my_function()
```

NULL

`function` will build a function out of whatever R code you place between the braces. For example, you can turn your dice code into a function by calling:

```
roll <- function() {
  die <- 1:6
  dice <- sample(die, size = 2, replace = TRUE)
  sum(dice)
}
```

Indentation and readability

Notice each line of code between the braces is indented. This makes the code easier to read but has no impact

3. Up and Running with R

on how the code runs. R ignores spaces and line breaks and executes one complete expression at a time. Note that in other languages like python, spacing is extremely important and part of the language.

Just hit the Enter key between each line after the first brace, {. R will wait for you to type the last brace, }, before it responds.

Don't forget to save the output of `function` to an R object. This object will become your new function. To use it, write the object's name followed by an open and closed parenthesis:

```
roll()
```

```
[1] 6
```

You can think of the parentheses as the “trigger” that causes R to run the function. If you type in a function's name *without* the parentheses, R will show you the code that is stored inside the function. If you type in the name *with* the parentheses, R will run that code:

```
roll
```

```
function() {  
  die <- 1:6  
  dice <- sample(die, size = 2, replace = TRUE)  
  sum(dice)  
}
```

```
roll()
```

```
[1] 6
```

The code that you place inside your function is known as the *body* of the function. When you run a function in R, R will execute all of the code in the body and then return the result

3. Up and Running with R

of the last line of code. If the last line of code doesn't return a value, neither will your function, so you want to ensure that your final line of code returns a value. One way to check this is to think about what would happen if you ran the body of code line by line in the command line. Would R display a result after the last line, or would it not?

Here's some code that would display a result:

```
dice  
1 + 1  
sqrt(2)
```

And here's some code that would not:

```
dice <- sample(die, size = 2, replace = TRUE)  
two <- 1 + 1  
a <- sqrt(2)
```

Again, this is just showing the distinction between expressions and assignments.

3.5. Arguments

What if we removed one line of code from our function and changed the name `die` to `bones` (just a name—don't think of it as important), like this?

```
roll2 <- function() {  
  dice <- sample(bones, size = 2, replace = TRUE)  
  sum(dice)  
}
```

Now I'll get an error when I run the function. The function **needs** the object `bones` to do its job, but there is no object named `bones` to be found (you can check by typing `ls()` which will show you the names in the environment, or memory).

3. Up and Running with R

```
roll2()
## Error in sample(bones, size = 2, replace = TRUE) :
##   object 'bones' not found
```

You can supply `bones` when you call `roll2` if you make `bones` an argument of the function. To do this, put the name `bones` in the parentheses that follow `function` when you define `roll2`:

```
roll2 <- function(bones) {
  dice <- sample(bones, size = 2, replace = TRUE)
  sum(dice)
}
```

Now `roll2` will work as long as you supply `bones` when you call the function. You can take advantage of this to roll different types of dice each time you call `roll2`.

Remember, we're rolling pairs of dice:

```
roll2(bones = 1:4)
```

```
[1] 4
```

```
roll2(bones = 1:6)
```

```
[1] 5
```

```
roll2(1:20)
```

```
[1] 23
```

Notice that `roll2` will still give an error if you do not supply a value for the `bones` argument when you call `roll2`:

```
roll2()
## Error in sample(bones, size = 2, replace = TRUE) :
##   argument "bones" is missing, with no default
```

3. Up and Running with R

You can prevent this error by giving the `bones` argument a default value. To do this, set `bones` equal to a value when you define `roll2`:

```
roll2 <- function(bones = 1:6) {  
  dice <- sample(bones, size = 2, replace = TRUE)  
  sum(dice)  
}
```

Now you can supply a new value for `bones` if you like, and `roll2` will use the default if you do not:

```
roll2()
```

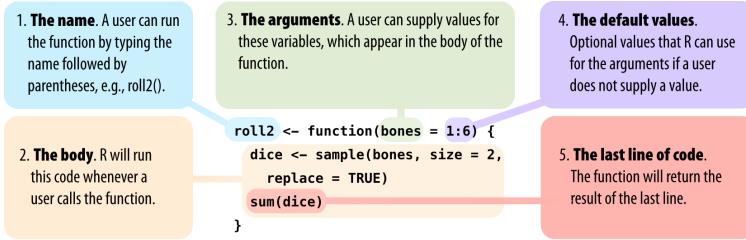
```
[1] 6
```

You can give your functions as many arguments as you like. Just list their names, separated by commas, in the parentheses that follow `function`. When the function is run, R will replace each argument name in the function body with the value that the user supplies for the argument. If the user does not supply a value, R will replace the argument name with the argument's default value (if you defined one).

To summarize, `function` helps you construct your own R functions. You create a body of code for your function to run by writing code between the braces that follow `function`. You create arguments for your function to use by supplying their names in the parentheses that follow `function`. Finally, you give your function a name by saving its output to an R object, as shown in Figure 3.6.

Once you've created your function, R will treat it like every other function in R. Think about how useful this is. Have you ever tried to create a new Excel option and add it to Microsoft's menu bar? Or a new slide animation and add it to Powerpoint's options? When you work with a programming language, you can do these types of things. As you learn to program in R, you will be able to create new, customized, reproducible tools for yourself whenever you like.

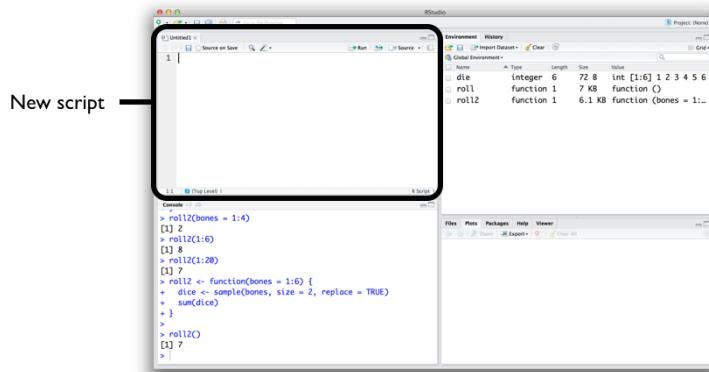
3. Up and Running with R



3.6. Scripts

Scripts are code that are saved for later reuse or editing. An R script is just a plain text file that you save R code in. You can open an R script in RStudio by going to **File > New File > R script** in the menu bar. RStudio will then open a fresh script above your console pane, as shown in Figure 3.7.

I strongly encourage you to write and edit all of your R code in a script before you run it in the console. Why? This habit creates a reproducible record of your work. When you're finished for the day, you can save your script and then use it to rerun your entire analysis the next day. Scripts are also very handy for editing and proofreading your code, and they make a nice copy of your work to share with others. To save a script, click the scripts pane, and then go to **File > Save As** in the menu bar.



RStudio comes with many built-in features that make it easy

Figure 3.6.: “Every function in R has the same parts, and you can use `function` to create these parts. Assign the result to a name, so you can call the function later.”

Figure 3.7.: “When you open an R Script (File > New File > R Script in the menu bar), RStudio creates a fourth pane (or puts a new tab in the existing pane) above the console where you can write and edit your code.”

3. Up and Running with R

to work with scripts. First, you can automatically execute a line of code in a script by clicking the Run button at the top of the editor panel.

R will run whichever line of code your cursor is on. If you have a whole section highlighted, R will run the highlighted code. Alternatively, you can run the entire script by clicking the Source button. Don't like clicking buttons? You can use Control + Return as a shortcut for the Run button. On Macs, that would be Command + Return.

If you're not convinced about scripts, you soon will be. It becomes a pain to write multi-line code in the console's single-line command line. Let's avoid that headache and open your first script now before we move to the next chapter.

💡 Tip

Extract function

RStudio comes with a tool that can help you build functions. To use it, highlight the lines of code in your R script that you want to turn into a function. Then click **Code > Extract Function** in the menu bar. RStudio will ask you for a function name to use and then wrap your code in a **function** call. It will scan the code for undefined variables and use these as arguments.

You may want to double-check RStudio's work. It assumes that your code is correct, so if it does something surprising, you may have a problem in your code.

3.7. Summary

We've covered a lot of ground already. You now have a virtual die stored in your computer's memory, as well as your own R function that rolls a pair of dice. You've also begun speaking the R language.

The two most important components of the R language are objects, which store data, and functions, which manipulate data. R also uses a host of operators like `+`, `-`, `*`, `/`, and `<-` to do basic tasks. As a data scientist, you will use R objects to store

3. Up and Running with R

data in your computer’s memory, and you will use functions to automate tasks and do complicated calculations.

4. Packages and more dice

We now have code that allows us to roll two dice and add the results together. To keep things interesting, let's aim to weight the dice so that we can fool our friends into thinking we are lucky.

First, though, we should prove to ourselves that our dice are fair. We can investigate the behavior of our dice using two powerful and general tools;

- Simulation (or repetition or repeated sampling)
- Visualization

For the repetition part of things, we will use a built-in R function, `replicate`. For visualization, we are going to use a convenient plotting function, `qplot`. However, `qplot` does not come built into R. We must install a *package* to gain access to it.

4.1. Packages

R is a powerful language for data science and programming, allowing beginners and experts alike to manipulate, analyze, and visualize data effectively. One of the most appealing features of R is its extensive library of packages, which are essential tools for expanding its capabilities and streamlining the coding process.

An R package is a collection of reusable functions, datasets, and compiled code created by other users and developers to extend the functionality of the base R language. These packages cover a wide range of applications, such as data manipulation, statistical analysis, machine learning, and data visualization. By utilizing existing R packages, you can leverage the expertise

4. Packages and more dice

of others and save time by avoiding the need to create custom functions from scratch.

Using others' R packages is incredibly beneficial as it allows you to take advantage of the collective knowledge of the R community. Developers often create packages to address specific challenges, optimize performance, or implement popular algorithms or methodologies. By incorporating these packages into your projects, you can enhance your productivity, reduce development time, and ensure that you are using well-tested and reliable code.

4.1.1. `install.packages`

To install an R package, you can use the `install.packages()` function in the R console or script. For example, to install the popular data manipulation package “dplyr,” simply type `install.packages("dplyr")`. This command will download the package from the Comprehensive R Archive Network (CRAN) and install it on your local machine. Keep in mind that you only need to install a package once, unless you want to update it to a newer version.

In our case, we want to install the `ggplot2` package.

```
install.packages('ggplot2')
```

4.1.2. `library`

After installing an R package, you will need to load it into your R session before using its functions. To load a package, use the `library()` function followed by the package name, such as `library(dplyr)`. Loading a package makes its functions and datasets available for use in your current R session. Note that you need to load a package every time you start a new R session.

```
library(ggplot2)
```

4. Packages and more dice

Now, the functionality of the *ggplot2* package is available in our R session.

💡 Installing vs loading packages

The main thing to remember is that you only need to install a package once, but you need to load it with library each time you wish to use it in a new R session. R will unload all of its packages each time you close RStudio.

4.1.3. Finding R packages

Finding useful R packages can be done in several ways. First, browsing CRAN (<https://cran.r-project.org/>) and Bioconductor (more later, <https://bioconductor.org>) are an excellent starting points, as they host thousands of packages categorized by topic. Additionally, online forums like Stack Overflow and R-bloggers can provide valuable recommendations based on user experiences. Social media platforms such as Twitter, where developers and data scientists often share new packages and updates, can also be a helpful resource. Finally, don't forget to ask your colleagues or fellow R users for their favorite packages, as they may have insights on which ones best suit your specific needs.

4.2. Are our dice fair?

Well, let's review our code.

```
roll2 <- function(bones = 1:6) {  
  dice = sample(bones, size = 2, replace = TRUE)  
  sum(dice)  
}
```

If our dice are fair, then each number should show up equally. What does the sum look like with our two dice?

Read the help page for `replicate` (i.e., `help("replicate")`). In short, it suggests that we can repeat our dice rolling as many

4. Packages and more dice

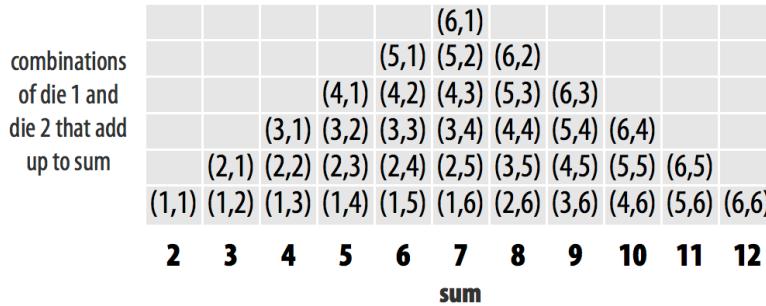


Figure 4.1.: In an ideal world, a histogram of the results would look like this

times as we like and replicate will return a *vector* of the sums for each roll.

```
rolls = replicate(n = 100, roll2())
```

What does rolls look like?

```
head(rolls)
```

```
[1] 11 10 5 9 6 7
```

```
length(rolls)
```

```
[1] 100
```

```
mean(rolls)
```

```
[1] 7.05
```

```
summary(rolls)
```

Min.	1st Qu.	Median	Mean	3rd Qu.	Max.
2.00	6.00	7.00	7.05	9.00	11.00

4. Packages and more dice

This looks like it roughly agrees with our sketched out ideal histogram in Figure 4.1. However, now that we've loaded the `qplot` function from the `ggplot2` package, we can make a histogram of the data themselves.

```
qplot(rolls, binwidth=1)
```

Warning: `qplot()` was deprecated in ggplot2 3.4.0.

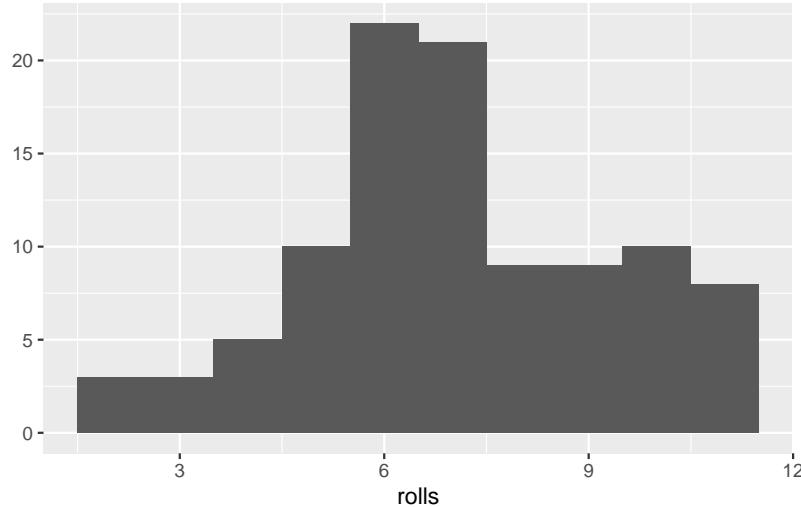


Figure 4.2.: Histogram of the sums from 100 rolls of our fair dice

How does your histogram look (and yours will be different from mine since we are sampling random values)? Is it what you expect?

What happens to our histogram as we increase the number of replicates?

```
rolls = replicate(n = 100000, roll2())
qplot(rolls, binwidth=1)
```

4. Packages and more dice

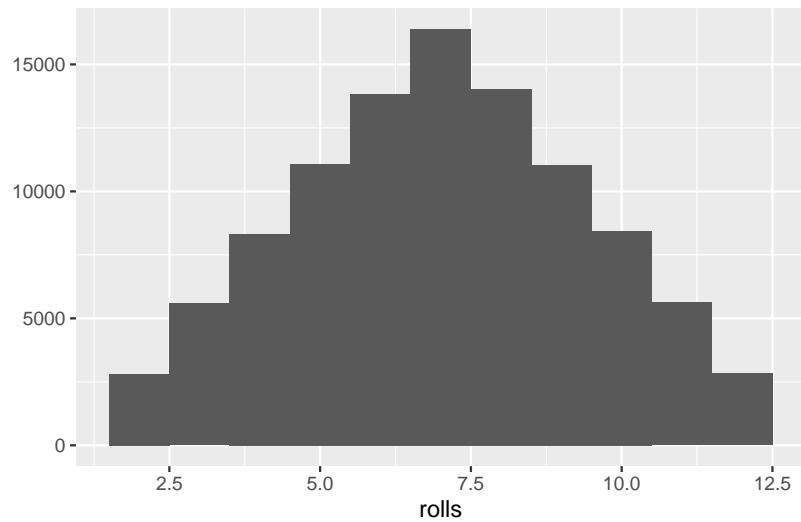


Figure 4.3.: Histogram with 100000 rolls much more closely approximates the pyramidal shape we anticipated

4.3. Bonus exercise

How would you change the `roll12` function to weight the dice?

5. Reading and writing data files

5.1. Introduction

In this chapter, we will discuss how to read and write data files in R. Data files are essential for storing and sharing data across different platforms and applications. R provides a variety of functions and packages to read and write data files in different formats, such as text files, CSV files, Excel files. By mastering these functions, you can efficiently import and export data in R, enabling you to perform data analysis and visualization tasks effectively.

5.2. CSV files

Comma-Separated Values (CSV) files are a common file format for storing tabular data. They consist of rows and columns, with each row representing a record and each column representing a variable or attribute. CSV files are widely used for data storage and exchange due to their simplicity and compatibility with various software applications. In R, you can read and write CSV files using the `read.csv()` and `write.csv()` functions, respectively. A commonly used alternative is to use the `readr` package, which provides faster and more user-friendly functions for reading and writing CSV files.

5.2.1. Writing a CSV file

Since we are going to use the `readr` package, we need to install it first. You can install the `readr` package using the following command:

5. Reading and writing data files

```
install.packages("readr")
```

Once the package is installed, you can load it into your R session using the `library()` function:

```
library(readr)
```

Since we don't have a CSV file sitting around, let's create a simple data frame to write to a CSV file. Here's an example data frame:

```
df <- data.frame(  
  id = c(1, 2, 3, 4, 5),  
  name = c("Alice", "Bob", "Charlie", "David", "Eve"),  
  age = c(25, 30, 35, 40, 45)  
)
```

Now, you can write this data frame to a CSV file using the `write_csv()` function from the `readr` package. Here's how you can do it:

```
write_csv(df, "data.csv")
```

You can check the current working directory to see if the CSV file was created successfully. If you want to specify a different directory or file path, you can provide the full path in the `write_csv()` function.

```
# see what the current working directory is  
getwd()
```

```
[1] "/Users/seandavis/Documents/git/RBiocBook"
```

```
# and check to see that the file was created  
dir(pattern = "data.csv")
```

```
[1] "data.csv"
```

5. Reading and writing data files

5.2.2. Reading a CSV file

Now that we have a CSV file, let's read it back into R using the `read_csv()` function from the `readr` package. Here's how you can do it:

```
df2 <- read_csv("data.csv")  
  
Rows: 5 Columns: 3  
-- Column specification -----  
Delimiter: ","  
chr (1): name  
dbl (2): id, age  
  
i Use `spec()` to retrieve the full column specification for this data.  
i Specify the column types or set `show_col_types = FALSE` to quiet this message.
```

You can check the structure of the data frame `df2` to verify that the data was read correctly:

```
df2  
  
# A tibble: 5 x 3  
  id name     age  
  <dbl> <chr>   <dbl>  
1     1 Alice     25  
2     2 Bob       30  
3     3 Charlie   35  
4     4 David    40  
5     5 Eve      45
```

The `readr` package can read CSV files with various delimiters, headers, and data types, making it a versatile tool for handling tabular data in R. It can also read CSV files directly from web locations like so:

```
df3 <- read_csv("https://data.cdc.gov/resource/pwn4-m3yp.csv")
```

5. Reading and writing data files

```
Rows: 1000 Columns: 10
-- Column specification -----
Delimiter: ","
chr (1): state
dbl (6): tot_cases, new_cases, tot_deaths, new_deaths, new_historic_cases, ...
dttm (3): date_updated, start_date, end_date

i Use `spec()` to retrieve the full column specification for this data.
i Specify the column types or set `show_col_types = FALSE` to quiet this message.
```

The dataset that you just downloaded is described here: [Covid-19 data from CDC](#)

5.3. Excel files

Microsoft Excel files are another common file format for storing tabular data. Excel files can contain multiple sheets, formulas, and formatting options, making them a popular choice for data storage and analysis. In R, you can read and write Excel files using the `readxl` package. This package provides functions to import and export data from Excel files, enabling you to work with Excel data in R.

5.3.1. Reading an Excel file

To read an Excel file in R, you need to install and load the `readxl` package. You can install the `readxl` package using the following command:

```
install.packages("readxl")
```

Once the package is installed, you can load it into your R session using the `library()` function:

```
library(readxl)
```

5. Reading and writing data files

Now, you can read an Excel file using the `read_excel()` function from the `readxl` package. We don't have an excel file available, so let's download one from the internet. Here's an example:

```
download.file('https://www.w3resource.com/python-exercises/pandas/excel/SaleData.xlsx', 'SaleD
```

Now, you can read the Excel file into R using the `read_excel()` function:

```
df_excel <- read_excel("SaleData.xlsx")
```

You can check the structure of the data frame `df_excel` to verify that the data was read correctly:

```
df_excel
```

```
# A tibble: 45 x 8
  OrderDate      Region Manager SalesMan Item  Units Unit_price Sale_amt
  <dttm>        <chr>   <chr>   <chr>   <chr> <dbl>    <dbl>    <dbl>
1 2018-01-06 00:00:00 East    Martha  Alexander Tele~    95     1198    113810
2 2018-01-23 00:00:00 Central Hermann Shelli   Home~    50      500    25000
3 2018-02-09 00:00:00 Central Hermann Luis     Tele~    36     1198    43128
4 2018-02-26 00:00:00 Central Timothy David   Cell~    27      225    6075
5 2018-03-15 00:00:00 West    Timothy Stephen  Tele~    56     1198    67088
6 2018-04-01 00:00:00 East    Martha  Alexander Home~    60      500    30000
7 2018-04-18 00:00:00 Central Martha  Steven   Tele~    75     1198    89850
8 2018-05-05 00:00:00 Central Hermann Luis     Tele~    90     1198    107820
9 2018-05-22 00:00:00 West    Douglas Michael  Tele~    32     1198    38336
10 2018-06-08 00:00:00 East   Martha  Alexander Home~    60      500    30000
# i 35 more rows
```

The `readxl` package provides various options to read Excel files with multiple sheets, specific ranges, and data types, making it a versatile tool for handling Excel data in R.

5. Reading and writing data files

5.3.2. Writing an Excel file

To write an Excel file in R, you can use the `write_xlsx()` function from the `writexl` package. You can install the `writexl` package using the following command:

```
install.packages("writexl")
```

Once the package is installed, you can load it into your R session using the `library()` function:

```
library(writexl)
```

The `write_xlsx()` function allows you to write a data frame to an Excel file. Here's an example:

```
write_xlsx(df, "data.xlsx")
```

You can check the current working directory to see if the Excel file was created successfully. If you want to specify a different directory or file path, you can provide the full path in the `write_xlsx()` function.

```
# see what the current working directory is  
getwd()
```

```
[1] "/Users/seandavis/Documents/git/RBiocBook"
```

```
# and check to see that the file was created  
dir(pattern = "data.xlsx")
```

```
[1] "data.xlsx"
```

5. Reading and writing data files

5.4. Additional options

- Google Sheets: You can read and write data from Google Sheets using the `googlesheets4` package. This package provides functions to interact with Google Sheets, enabling you to import and export data from Google Sheets to R.
- JSON files: You can read and write JSON files using the `jsonlite` package. This package provides functions to convert R objects to JSON format and vice versa, enabling you to work with JSON data in R.
- Database files: You can read and write data from database files using the `DBI` and `RSQLite` packages. These packages provide functions to interact with various database systems, enabling you to import and export data from databases to R.

Part II.

R Data Structures

Welcome to the section on R data structures! As you begin your journey in learning R, it is essential to understand the fundamental building blocks of this powerful programming language. R offers a variety of data structures to store and manipulate data, each with its unique properties and capabilities. In this section, we will cover the core data structures in R, including:

- Vectors
- Matrices
- Lists
- Data.frames

By the end of this section, you will have a solid understanding of these data structures, and you will be able to choose and utilize the appropriate data structure for your specific data manipulation and analysis tasks.

In each chapter, we will delve into the properties and usage of each data structure, starting with their definitions and moving on to their practical applications. We will provide examples, exercises, and active learning approaches to help you better understand and apply these concepts in your work.

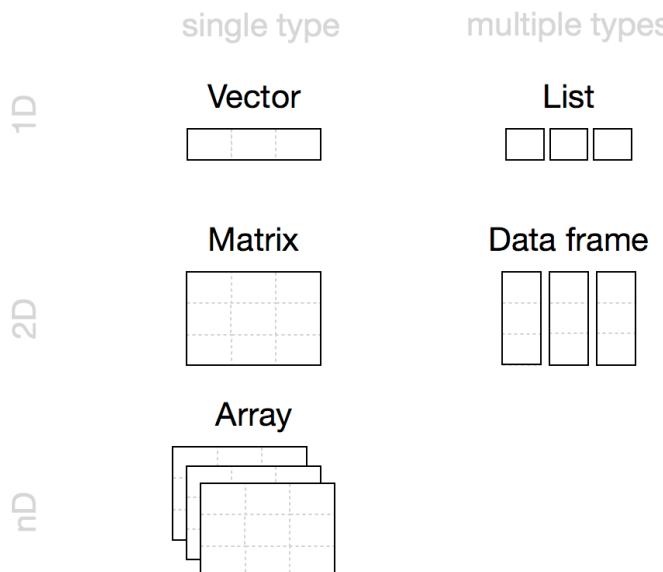


Figure 5.1.: A pictorial representation of R's most common data structures are vectors, matrices, arrays, lists, and dataframes. Figure from [Hands-on Programming with R](#).

Chapter overview

- **Vectors** : In this chapter, we will introduce you to the simplest data structure in R, the vector. We will cover how to create, access, and manipulate vectors, as well as discuss their unique properties and limitations.
- **Matrices** Next, we will explore matrices, which are two-dimensional data structures that extend vectors. You will learn how to create, access, and manipulate matrices, and understand their usefulness in mathematical operations and data organization.
- **Lists** The third chapter will focus on lists, a versatile data structure that can store elements of different types and sizes. We will discuss how to create, access, and modify lists, and demonstrate their flexibility in handling complex data structures.
- **Data.frames** Finally, we will examine data.frames, a widely-used data structure for organizing and manipulating tabular data. You will learn how to create, access, and manipulate data.frames, and understand their advantages over other data structures for data analysis tasks.
- **Arrays** While we will not focus directly on the `array` data type, which are multidimensional data structures that extend matrices, they are very similar to matrices, but with a third dimension.

As you progress through these chapters, practice the examples and exercises provided, engage in discussion, and collaborate with your peers to deepen your understanding of R data structures. This solid foundation will serve as the basis for more advanced data manipulation, analysis, and visualization techniques in R.

6. Vectors

6.1. What is a Vector?

A vector is the simplest and most basic data structure in R. It is a one-dimensional, ordered collection of elements, where all the elements are of the same data type. Vectors can store various types of data, such as numeric, character, or logical values. Figure 6.1 shows a pictorial representation of three vector examples.

Index	1	2	3	4	5	6	7
Vector	3	7	10	NA	932	127	-3
Vector	TRUE	FALSE	FALSE	TRUE	TRUE	FALSE	NA
Vector	“Cat”	“Dog”	“A”	“C”	“T”	NA	“G”
Names (Optional)	“H”	“I”	“L”	“Z”	“This”	“That”	“Other”

In this chapter, we will provide a comprehensive overview of vectors, including how to create, access, and manipulate them. We will also discuss some unique properties and rules associated with vectors, and explore their applications in data analysis tasks.

In R, even a single value is a vector with length=1.

```
z = 1
z
```

```
[1] 1
```

Figure 6.1.: “Pictorial representation of three vector examples. The first vector is a numeric vector. The second is a ‘logical’ vector. The third is a character vector. Vectors also have indices and, optionally, names.”

6. Vectors

```
length(z)
```

```
[1] 1
```

In the code above, we “assigned” the value 1 to the variable named `z`. Typing `z` by itself is an “expression” that returns a result which is, in this case, the value that we just assigned. The `length` method takes an R object and returns the R length. There are numerous ways of asking R about what an object represents, and `length` is one of them.

Vectors can contain numbers, strings (character data), or logical values (`TRUE` and `FALSE`) or other “atomic” data types Table 6.1. *Vectors cannot contain a mix of types!* We will introduce another data structure, the R `list` for situations when we need to store a mix of base R data types.

Table 6.1.: Atomic (simplest) data types in R.

Data type	Stores
numeric	floating point numbers
integer	integers
complex	complex numbers
factor	categorical data
character	strings
logical	TRUE or FALSE
NA	missing
NULL	empty
function	function type

6.2. Creating vectors

Character vectors (also sometimes called “string” vectors) are entered with each value surrounded by single or double quotes; either is acceptable, but they must match. They are always displayed by R with double quotes. Here are some examples of creating vectors:

6. Vectors

```
# examples of vectors
c('hello','world')

[1] "hello" "world"

c(1,3,4,5,1,2)

[1] 1 3 4 5 1 2

c(1.12341e7,78234.126)

[1] 11234100.00    78234.13

c(TRUE,FALSE,TRUE,TRUE)

[1] TRUE FALSE  TRUE  TRUE

# note how in the next case the TRUE is converted to "TRUE"
# with quotes around it.
c(TRUE,'hello')

[1] "TRUE"  "hello"
```

We can also create vectors as “regular sequences” of numbers.
For example:

```
# create a vector of integers from 1 to 10
x = 1:10
# and backwards
x = 10:1
```

The `seq` function can create more flexible regular sequences.

```
# create a vector of numbers from 1 to 4 skipping by 0.3
y = seq(1,4,0.3)
```

And creating a new vector by concatenating existing vectors is possible, as well.

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```
# create a sequence by concatenating two other sequences
z = c(y,x)
z
```

```
[1] 1.0 1.3 1.6 1.9 2.2 2.5 2.8 3.1 3.4 3.7 4.0 10.0 9.0 8.0 7.0
[16] 6.0 5.0 4.0 3.0 2.0 1.0
```

6.3. Vector Operations

Operations on a single vector are typically done element-by-element. For example, we can add 2 to a vector, 2 is added to each element of the vector and a new vector of the same length is returned.

```
x = 1:10
x + 2
```

```
[1] 3 4 5 6 7 8 9 10 11 12
```

If the operation involves two vectors, the following rules apply. If the vectors are the same length: R simply applies the operation to each pair of elements.

```
x + x
```

```
[1] 2 4 6 8 10 12 14 16 18 20
```

If the vectors are different lengths, but one length a multiple of the other, R reuses the shorter vector as needed.

```
x = 1:10
y = c(1,2)
x * y
```

```
[1] 1 4 3 8 5 12 7 16 9 20
```

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If the vectors are different lengths, but one length *not* a multiple of the other, R reuses the shorter vector as needed *and* delivers a warning.

```
x = 1:10  
y = c(2,3,4)  
x * y
```

```
Warning in x * y: longer object length is not a multiple of shorter object  
length
```

```
[1] 2 6 12 8 15 24 14 24 36 20
```

Typical operations include multiplication (“*”), addition, subtraction, division, exponentiation (“^”), but many operations in R operate on vectors and are then called “vectorized”.

Be aware of the recycling rule when working with vectors of different lengths, as it may lead to unexpected results if you’re not careful.

6.4. Logical Vectors

Logical vectors are vectors composed on only the values TRUE and FALSE. Note the all-upper-case and no quotation marks.

```
a = c(TRUE, FALSE, TRUE)  
  
# we can also create a logical vector from a numeric vector  
# 0 = false, everything else is 1  
b = c(1, 0, 217)  
d = as.logical(b)  
d
```

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

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```
# test if a and d are the same at every element  
all.equal(a,d)
```

```
[1] TRUE
```

```
# We can also convert from logical to numeric  
as.numeric(a)
```

```
[1] 1 0 1
```

6.4.1. Logical Operators

Some operators like `<`, `>`, `==`, `>=`, `<=`, `!=` can be used to create logical vectors.

```
# create a numeric vector  
x = 1:10  
# testing whether x > 5 creates a logical vector  
x > 5
```

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

```
x <= 5
```

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

```
x != 5
```

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

```
x == 5
```

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

We can also assign the results to a variable:

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```
y = (x == 5)  
y
```

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

6.5. Indexing Vectors

In R, an index is used to refer to a specific element or set of elements in a vector (or other data structure). [R uses [and] to perform indexing, although other approaches to getting subsets of larger data structures are common in R.

```
x = seq(0,1,0.1)  
# create a new vector from the 4th element of x  
x[4]
```

```
[1] 0.3
```

We can even use other vectors to perform the “indexing”.

```
x[c(3,5,6)]
```

```
[1] 0.2 0.4 0.5
```

```
y = 3:6  
x[y]
```

```
[1] 0.2 0.3 0.4 0.5
```

Combining the concept of indexing with the concept of logical vectors results in a very power combination.

```
# use help('rnorm') to figure out what is happening next  
myvec = rnorm(10)  
  
# create logical vector that is TRUE where myvec is >0.25  
gt1 = (myvec > 0.25)  
sum(gt1)
```

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```
[1] 4
```

```
# and use our logical vector to create a vector of myvec values that are >0.25  
myvec[gt1]
```

```
[1] 1.1484509 1.1463211 0.7716711 0.2969809
```

```
# or <=0.25 using the logical "not" operator, "!"  
myvec[!gt1]
```

```
[1] -0.4014349 -0.5081373 -0.4925580 -1.6429488 -0.1851662 -1.0668761
```

```
# shorter, one line approach  
myvec[myvec > 0.25]
```

```
[1] 1.1484509 1.1463211 0.7716711 0.2969809
```

6.6. Named Vectors

Named vectors are vectors with labels or names assigned to their elements. These names can be used to access and manipulate the elements in a more meaningful way.

To create a named vector, use the `names()` function:

```
fruit_prices <- c(0.5, 0.75, 1.25)  
names(fruit_prices) <- c("apple", "banana", "cherry")  
print(fruit_prices)
```

```
apple banana cherry  
0.50   0.75   1.25
```

You can also access and modify elements using their names:

```
banana_price <- fruit_prices["banana"]  
print(banana_price)
```

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```
banana
0.75

fruit_prices["apple"] <- 0.6
print(fruit_prices)

apple banana cherry
0.60   0.75   1.25
```

6.7. Character Vectors, A.K.A. Strings

R uses the `paste` function to concatenate strings.

```
paste("abc", "def")

[1] "abc def"

paste("abc", "def", sep="THISSEP")

[1] "abcTHISSEPdef"

paste0("abc", "def")

[1] "abcdef"

## [1] "abcdef"
paste(c("X", "Y"), 1:10)

[1] "X 1"   "Y 2"   "X 3"   "Y 4"   "X 5"   "Y 6"   "X 7"   "Y 8"   "X 9"   "Y 10"

paste(c("X", "Y"), 1:10, sep="_")

[1] "X_1"   "Y_2"   "X_3"   "Y_4"   "X_5"   "Y_6"   "X_7"   "Y_8"   "X_9"   "Y_10"
```

We can count the number of characters in a string.

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```
nchar('abc')
```

```
[1] 3
```

```
nchar(c('abc','d',123456))
```

```
[1] 3 1 6
```

Pulling out parts of strings is also sometimes useful.

```
substr('This is a good sentence.',start=10,stop=15)
```

```
[1] " good "
```

Another common operation is to replace something in a string with something (a find-and-replace).

```
sub('This','That','This is a good sentence.')
```

```
[1] "That is a good sentence."
```

When we want to find all strings that match some other string, we can use `grep`, or “grab regular expression”.

```
grep('bcd',c('abcdef','abcd','bcde','cdef','defg'))
```

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

```
grep('bcd',c('abcdef','abcd','bcde','cdef','defg')),value=TRUE)
```

```
[1] "abcdef" "abcd"   "bcde"
```

Read about the `grep1` function (`?grep1`). Use that function to return a logical vector (TRUE/FALSE) for each entry above with an `a` in it.

6.8. Missing Values, AKA “NA”

R has a special value, “NA”, that represents a “missing” value, or *Not Available*, in a vector or other data structure. Here, we just create a vector to experiment.

```
x = 1:5
x
```

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

```
length(x)
```

```
[1] 5
```

```
is.na(x)
```

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

```
x[2] = NA
x
```

```
[1] 1 NA 3 4 5
```

The length of `x` is unchanged, but there is one value that is marked as “missing” by virtue of being `NA`.

```
length(x)
```

```
[1] 5
```

```
is.na(x)
```

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

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We can remove NA values by using indexing. In the following, `is.na(x)` returns a logical vector the length of `x`. The `!` is the logical *NOT* operator and converts TRUE to FALSE and vice-versa.

```
x[!is.na(x)]
```

```
[1] 1 3 4 5
```

6.9. Exercises

1. Create a numeric vector called `temperatures` containing the following values: 72, 75, 78, 81, 76, 73.

```
temperatures <- c(72, 75, 78, 81, 76, 73, 93)
```

2. Create a character vector called `days` containing the following values: “Monday”, “Tuesday”, “Wednesday”, “Thursday”, “Friday”, “Saturday”, “Sunday”.

```
days <- c("Monday", "Tuesday", "Wednesday", "Thursday", "Friday", "Saturday", "Sunday")
```

3. Calculate the average temperature for the week and store it in a variable called `average_temperature`.

```
average_temperature <- mean(temperatures)
```

4. Create a named vector called `weekly_temperatures`, where the names are the days of the week and the values are the temperatures from the `temperatures` vector.

```
weekly_temperatures <- temperatures  
names(weekly_temperatures) <- days
```

5. Create a numeric vector called `ages` containing the following values: 25, 30, 35, 40, 45, 50, 55, 60.

```
ages <- c(25, 30, 35, 40, 45, 50, 55, 60)
```

6. Create a logical vector called `is_adult` by checking if the elements in the `ages` vector are greater than or equal to 18.

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```
is_adult <- ages >= 18
```

7. Calculate the sum and product of the `ages` vector.

```
sum_ages <- sum(ages)  
product_ages <- prod(ages)
```

8. Extract the ages greater than or equal to 40 from the `ages` vector and store them in a variable called `older_ages`.

```
older_ages <- ages[ages >= 40]
```

7. Matrices

A *matrix* is a rectangular collection of the same data type (see Figure 7.1). It can be viewed as a collection of column vectors all of the same length and the same type (i.e. numeric, character or logical) OR a collection of row vectors, again all of the same type and length. A *data.frame* is *also* a rectangular array. All of the columns must be the same length, but they **may be** of *different* types. The rows and columns of a matrix or data frame can be given names. However these are implemented differently in R; many operations will work for one but not both, often a source of confusion.

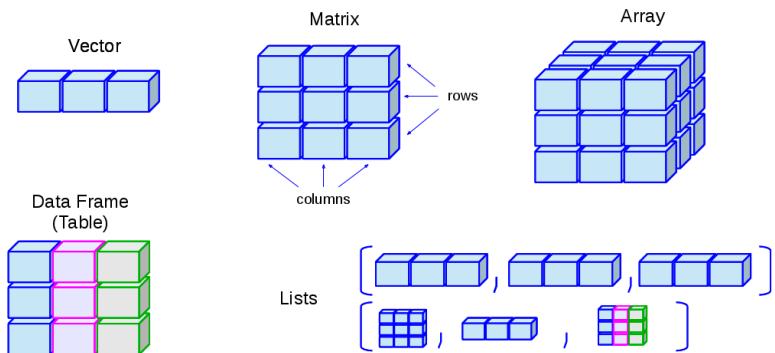


Figure 7.1.: A matrix is a collection of column vectors.

7.1. Creating a matrix

There are many ways to create a matrix in R. One of the simplest is to use the `matrix()` function. In the code below, we'll create a matrix from a vector from 1:16.

```
mat1 <- matrix(1:16, nrow=4)
mat1
```

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```
[,1] [,2] [,3] [,4]
[1,]    1     5     9    13
[2,]    2     6    10    14
[3,]    3     7    11    15
[4,]    4     8    12    16
```

The same is possible, but specifying that the matrix be “filled” by row.

```
mat1 <- matrix(1:16,nrow=4,byrow = TRUE)
mat1
```

```
[,1] [,2] [,3] [,4]
[1,]    1     2     3     4
[2,]    5     6     7     8
[3,]    9    10    11    12
[4,]   13    14    15    16
```

Notice the subtle difference in the order that the numbers go into the matrix.

We can also build a matrix from parts by “binding” vectors together:

```
x <- 1:10
y <- rnorm(10)
```

Each of the vectors above is of length 10 and both are “numeric”, so we can make them into a matrix. Using `rbind` binds rows (`r`) into a matrix.

```
mat <- rbind(x,y)
mat
```

```
[,1]      [,2]      [,3]      [,4]      [,5]      [,6]      [,7]
x  1.0000000 2.0000000 3.0000000 4.0000000 5.0000000 6.000000 7.0000000
y -0.1007675 0.5519366 0.4488688 0.3981466 0.8524107 -1.027999 -0.6854053
[,8]      [,9]      [,10]
x 8.0000000 9.0000000 10.0000000
y 0.4897315 -0.2333974 0.7278752
```

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The alternative to `rbind` is `cbind` that binds columns (**c**) together.

```
mat <- cbind(x,y)
mat
```

```
      x          y
[1,] 1 -0.1007675
[2,] 2  0.5519366
[3,] 3  0.4488688
[4,] 4  0.3981466
[5,] 5  0.8524107
[6,] 6 -1.0279989
[7,] 7 -0.6854053
[8,] 8  0.4897315
[9,] 9 -0.2333974
[10,] 10 0.7278752
```

Inspecting the names associated with rows and columns is often useful, particularly if the names have human meaning.

```
rownames(mat)
```

```
NULL
```

```
colnames(mat)
```

```
[1] "x" "y"
```

We can also change the names of the matrix by assigning *valid* names to the columns or rows.

```
colnames(mat) = c('apples','oranges')
colnames(mat)
```

```
[1] "apples"   "oranges"
```

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```
mat
```

```
    apples      oranges
[1,]      1 -0.1007675
[2,]      2  0.5519366
[3,]      3  0.4488688
[4,]      4  0.3981466
[5,]      5  0.8524107
[6,]      6 -1.0279989
[7,]      7 -0.6854053
[8,]      8  0.4897315
[9,]      9 -0.2333974
[10,]     10  0.7278752
```

Matrices have dimensions.

```
dim(mat)
```

```
[1] 10  2
```

```
nrow(mat)
```

```
[1] 10
```

```
ncol(mat)
```

```
[1] 2
```

7.2. Accessing elements of a matrix

Indexing for matrices works as for vectors except that we now need to include both the row and column (in that order). We can access elements of a matrix using the square bracket [indexing method. Elements can be accessed as `var[r, c]`. Here, `r` and `c` are vectors describing the elements of the matrix to select.

7. Matrices

! Important

The indices in R start with one, meaning that the first element of a vector or the first row/column of a matrix is indexed as one.

This is different from some other programming languages, such as Python, which use zero-based indexing, meaning that the first element of a vector or the first row/column of a matrix is indexed as zero.

It is important to be aware of this difference when working with data in R, especially if you are coming from a programming background that uses zero-based indexing. Using the wrong index can lead to unexpected results or errors in your code.

```
# The 2nd element of the 1st row of mat  
mat[1,2]
```

```
oranges  
-0.1007675
```

```
# The first ROW of mat  
mat[1,]
```

```
apples      oranges  
1.0000000 -0.1007675
```

```
# The first COLUMN of mat  
mat[,1]
```

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

```
# and all elements of mat that are > 4; note no comma  
mat[mat>4]
```

```
[1] 5 6 7 8 9 10
```

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```
## [1] 5 6 7 8 9 10
```

🔥 Caution

Note that in the last case, there is no “,”, so R treats the matrix as a long vector (length=20). This is convenient, sometimes, but it can also be a source of error, as some code may “work” but be doing something unexpected.

We can also use indexing to exclude a row or column by prefixing the selection with a - sign.

```
mat[,-1]      # remove first column
```

```
[1] -0.1007675  0.5519366  0.4488688  0.3981466  0.8524107 -1.0279989  
[7] -0.6854053  0.4897315 -0.2333974  0.7278752
```

```
mat[-c(1:5),]  # remove first five rows
```

```
    apples     oranges  
[1,]       6 -1.0279989  
[2,]       7 -0.6854053  
[3,]       8  0.4897315  
[4,]       9 -0.2333974  
[5,]      10  0.7278752
```

7.3. Changing values in a matrix

We can create a matrix filled with random values drawn from a normal distribution for our work below.

```
m = matrix(rnorm(20), nrow=10)  
summary(m)
```

```
          V1              V2  
Min.   :-2.1707   Min.   :-1.78021  
1st Qu.:-1.4913   1st Qu.:-0.68510
```

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```
Median :-0.1734 Median :-0.37670
Mean   :-0.1912 Mean   :-0.04895
3rd Qu.: 0.5173 3rd Qu.: 0.96281
Max.   : 2.6163 Max.   : 1.39484
```

Multiplication and division works similarly to vectors. When multiplying by a vector, for example, the values of the vector are reused. In the simplest case, let's multiply the matrix by a constant (vector of length 1).

```
# multiply all values in the matrix by 20
m2 = m*20
summary(m2)
```

V1	V2
Min. :-43.414	Min. :-35.604
1st Qu.:-29.826	1st Qu.:-13.702
Median : -3.467	Median : -7.534
Mean : -3.823	Mean : -0.979
3rd Qu.: 10.347	3rd Qu.: 19.256
Max. : 52.326	Max. : 27.897

By combining subsetting with assignment, we can make changes to just part of a matrix.

```
# and add 100 to the first column of m
m2[,1] = m2[,1] + 100
# summarize m
summary(m2)
```

V1	V2
Min. : 56.59	Min. :-35.604
1st Qu.: 70.17	1st Qu.:-13.702
Median : 96.53	Median : -7.534
Mean : 96.18	Mean : -0.979
3rd Qu.:110.35	3rd Qu.: 19.256
Max. :152.33	Max. : 27.897

7. Matrices

A somewhat common transformation for a matrix is to transpose which changes rows to columns. One might need to do this if an assay output from a lab machine puts samples in rows and genes in columns, for example, while in Bioconductor/R, we often want the samples in columns and the genes in rows.

```
t(m2)
```

```
[,1]      [,2]      [,3]      [,4]      [,5]      [,6]      [,7]
[1,] 76.265132 130.080007 68.14308 67.34310 152.32616 106.24387 56.58636
[2,] -5.879369 -9.188765 -18.58693 27.89672 20.71269 -35.60416 -13.89722
[,8]      [,9]      [,10]
[1,] 111.71414 102.8434 90.22191
[2,] 14.88633 -13.1163 22.98715
```

7.4. Calculations on matrix rows and columns

Again, we just need a matrix to play with. We'll use `rnorm` again, but with a slight twist.

```
m3 = matrix(rnorm(100,5,2),ncol=10) # what does the 5 mean here? And the 2?
```

Since these data are from a normal distribution, we can look at a row (or column) to see what the mean and standard deviation are.

```
mean(m3[,1])
```

```
[1] 5.434771
```

```
sd(m3[,1])
```

```
[1] 1.675129
```

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```
# or a row  
mean(m3[1,])
```

```
[1] 6.147223
```

```
sd(m3[1,])
```

```
[1] 1.630307
```

There are some useful convenience functions for computing means and sums of data in **all** of the columns and rows of matrices.

```
colMeans(m3)
```

```
[1] 5.434771 5.177531 5.179380 4.965027 4.933516 4.238210 5.186793 3.976971  
[9] 4.788226 4.295322
```

```
rowMeans(m3)
```

```
[1] 6.147223 3.438289 4.920728 5.254608 3.609042 5.730218 4.280746 4.563036  
[9] 5.325723 4.906131
```

```
rowSums(m3)
```

```
[1] 61.47223 34.38289 49.20728 52.54608 36.09042 57.30218 42.80746 45.63036  
[9] 53.25723 49.06131
```

```
colSums(m3)
```

```
[1] 54.34771 51.77531 51.79380 49.65027 49.33516 42.38210 51.86793 39.76971  
[9] 47.88226 42.95322
```

We can look at the distribution of column means:

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```
# save as a variable  
cmeans = colMeans(m3)  
summary(cmeans)
```

Min.	1st Qu.	Median	Mean	3rd Qu.	Max.
3.977	4.419	4.949	4.818	5.179	5.435

Note that this is centered pretty closely around the selected mean of 5 above.

How about the standard deviation? There is not a `colSd` function, but it turns out that we can easily apply functions that take vectors as input, like `sd` and “apply” them across either the rows (the first dimension) or columns (the second) dimension.

```
csds = apply(m3, 2, sd)  
summary(csds)
```

Min.	1st Qu.	Median	Mean	3rd Qu.	Max.
1.054	1.677	1.791	1.811	1.953	2.420

Again, take a look at the distribution which is centered quite close to the selected standard deviation when we created our matrix.

7.5. Exercises

7.5.1. Data preparation

For this set of exercises, we are going to rely on a dataset that comes with R. It gives the number of sunspots per month from 1749-1983. The dataset comes as a `ts` or time series data type which I convert to a matrix using the following code.

Just run the code as is and focus on the rest of the exercises.

7. Matrices

```
data(sunspots)
sunspot_mat <- matrix(as.vector(sunspots), ncol=12, byrow = TRUE)
colnames(sunspot_mat) <- as.character(1:12)
rownames(sunspot_mat) <- as.character(1749:1983)
```

7.5.2. Questions

- After the conversion above, what does `sunspot_mat` look like? Use functions to find the number of rows, the number of columns, the class, and some basic summary statistics.

```
ncol(sunspot_mat)
nrow(sunspot_mat)
dim(sunspot_mat)
summary(sunspot_mat)
head(sunspot_mat)
tail(sunspot_mat)
```

- Practice subsetting the matrix a bit by selecting:
 - The first 10 years (rows)
 - The month of July (7th column)
 - The value for July, 1979 using the rowname to do the selection.

```
sunspot_mat[1:10,]
sunspot_mat[,7]
sunspot_mat['1979',7]
```

- These next few exercises take advantage of the fact that calling a univariate statistical function (one that expects a vector) works for matrices by just making a vector of all the values in the matrix. What is the highest (`max`) number of sunspots recorded in these data?

```
max(sunspot_mat)
```

- And the minimum?

```
min(sunspot_mat)
```

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3. And the overall mean and median?

```
mean(sunspot_mat)  
median(sunspot_mat)
```

4. Use the `hist()` function to look at the distribution of all the monthly sunspot data.

```
hist(sunspot_mat)
```

5. Read about the `breaks` argument to `hist()` to try to increase the number of breaks in the histogram to increase the resolution slightly. Adjust your `hist()` and `breaks` to your liking.

```
hist(sunspot_mat, breaks=40)
```

6. Now, let's move on to summarizing the data a bit to learn about the pattern of sunspots varies by month or by year. Examine the dataset again. What do the columns represent? And the rows?

```
# just a quick glimpse of the data will give us a sense  
head(sunspot_mat)
```

7. We'd like to look at the distribution of sunspots by month. How can we do that?

```
# the mean of the columns is the mean number of sunspots per month.  
colMeans(sunspot_mat)  
  
# Another way to write the same thing:  
apply(sunspot_mat, 2, mean)
```

8. Assign the month summary above to a variable and summarize it to get a sense of the spread over months.

```
monthmeans = colMeans(sunspot_mat)  
summary(monthmeans)
```

9. Play the same game for years to get the per-year mean?

```
ymeans = rowMeans(sunspot_mat)  
summary(ymean)
```

10. Make a plot of the yearly means. Do you see a pattern?

7. Matrices

```
plot(ymean)
# or make it clearer
plot(ymean, type='l')
```

8. Data Frames

While R has many different data types, the one that is central to much of the power and popularity of R is the `data.frame`. A `data.frame` looks a bit like an R matrix in that it has two dimensions, rows and columns. However, `data.frames` are usually viewed as a set of columns representing variables and the rows representing the values of those variables. Importantly, a `data.frame` may contain *different* data types in each of its columns; matrices **must** contain only one data type. This distinction is important to remember, as there are *specific* approaches to working with R `data.frames` that may be different than those for working with matrices.

8.1. Learning goals

- Understand how `data.frames` are different from matrices.
- Know a few functions for examining the contents of a `data.frame`.
- List approaches for subsetting `data.frames`.
- Be able to load and save tabular data from and to disk.
- Show how to create a `data.frames` from scratch.

8.2. Learning objectives

- Load the yeast growth dataset into R using `read.csv`.
- Examine the contents of the dataset.
- Use subsetting to find genes that may be involved with nutrient metabolism and transport.
- Summarize data measurements by categories.

8. Data Frames

8.3. Dataset

The data used here are borrowed directly from the [fantastic Biocookbook tutorials](#) and are a cleaned up version of the data from [Brauer et al. Coordination of Growth Rate, Cell Cycle, Stress Response, and Metabolic Activity in Yeast \(2008\) Mol Biol Cell 19:352-367](#). These data are from a gene expression microarray, and in this paper the authors examine the relationship between growth rate and gene expression in yeast cultures limited by one of six different nutrients (glucose, leucine, ammonium, sulfate, phosphate, uracil). If you give yeast a rich media loaded with nutrients except restrict the supply of a single nutrient, you can control the growth rate to any rate you choose. By starving yeast of specific nutrients you can find genes that:

1. Raise or lower their expression in response to growth rate. Growth-rate dependent expression patterns can tell us a lot about cell cycle control, and how the cell responds to stress. The authors found that expression of >25% of all yeast genes is linearly correlated with growth rate, independent of the limiting nutrient. They also found that the subset of negatively growth-correlated genes is enriched for peroxisomal functions, and positively correlated genes mainly encode ribosomal functions.
2. Respond differently when different nutrients are being limited. If you see particular genes that respond very differently when a nutrient is sharply restricted, these genes might be involved in the transport or metabolism of that specific nutrient.

The dataset can be downloaded directly from:

- [brauer2007_tidy.csv](#)

We are going to read this dataset into R and then use it as a playground for learning about data.frames.

8.4. Reading in data

R has many capabilities for reading in data. Many of the functions have names that help us to understand what data format is to be expected. In this case, the filename that we want to read ends in `.csv`, meaning comma-separated-values. The `read.csv()` function reads in `.csv` files. As usual, it is worth reading `help('read.csv')` to get a better sense of the possible bells-and-whistles.

The `read.csv()` function can read directly from a URL, so we do not need to download the file directly. This dataset is relatively large (about 16MB), so this may take a bit depending on your network connection speed.

```
options(width=60)

url = paste0(
  'https://raw.githubusercontent.com',
  '/bioconnector/workshops/master/data/brauer2007_tidy.csv'
)
ydat <- read.csv(url)
```

Our variable, `ydat`, now “contains” the downloaded and read data. We can check to see what data type `read.csv` gave us:

```
class(ydat)
```

[1] "data.frame"

8.5. Inspecting data.frames

Our `ydat` variable is a `data.frame`. As I mentioned, the dataset is fairly large, so we will not be able to look at it all at once on the screen. However, R gives us many tools to inspect a `data.frame`.

- Overviews of content

8. Data Frames

- `head()` to show first few rows
- `tail()` to show last few rows
- Size
 - `dim()` for dimensions (rows, columns)
 - `nrow()`
 - `ncol()`
 - `object.size()` for power users interested in the memory used to store an object
- Data and attribute summaries
 - `colnames()` to get the names of the columns
 - `rownames()` to get the “names” of the rows—may not be present
 - `summary()` to get per-column summaries of the data in the `data.frame`.

```
head(ydat)
```

```
symbol systematic_name nutrient rate expression
1   SFB2          YNL049C  Glucose 0.05    -0.24
2   <NA>          YNL095C  Glucose 0.05     0.28
3   QRI7          YDL104C  Glucose 0.05    -0.02
4   CFT2          YLR115W  Glucose 0.05    -0.33
5   SS02          YMR183C  Glucose 0.05     0.05
6   PSP2          YML017W  Glucose 0.05    -0.69
                                bp
1       ER to Golgi transport
2   biological process unknown
3 proteolysis and peptidolysis
4      mRNA polyadenylation*
5          vesicle fusion*
6   biological process unknown
                                mf
1   molecular function unknown
2   molecular function unknown
3 metalloendopeptidase activity
4           RNA binding
5      t-SNARE activity
6   molecular function unknown
```

8. Data Frames

```
tail(ydat)
```

	symbol	systematic_name	nutrient	rate	expression
198425	DOA1	YKL213C	Uracil	0.3	0.14
198426	KRE1	YNL322C	Uracil	0.3	0.28
198427	MTL1	YGR023W	Uracil	0.3	0.27
198428	KRE9	YJL174W	Uracil	0.3	0.43
198429	UTH1	YKR042W	Uracil	0.3	0.19
198430	<NA>	YOL111C	Uracil	0.3	0.04
				bp	
198425		ubiquitin-dependent protein catabolism*			
198426		cell wall organization and biogenesis			
198427		cell wall organization and biogenesis			
198428		cell wall organization and biogenesis*			
198429		mitochondrion organization and biogenesis*			
198430		biological process unknown			
			mf		
198425		molecular function unknown			
198426		structural constituent of cell wall			
198427		molecular function unknown			
198428		molecular function unknown			
198429		molecular function unknown			
198430		molecular function unknown			

```
dim(ydat)
```

```
[1] 198430      7
```

```
nrow(ydat)
```

```
[1] 198430
```

```
ncol(ydat)
```

```
[1] 7
```

8. Data Frames

```
colnames(ydat)
```

```
[1] "symbol"           "systematic_name" "nutrient"  
[4] "rate"             "expression"       "bp"  
[7] "mf"
```

```
summary(ydat)
```

```
symbol          systematic_name      nutrient  
Length:198430    Length:198430      Length:198430  
Class :character Class :character   Class :character  
Mode  :character Mode  :character   Mode  :character
```

```
rate            expression          bp  
Min.  :0.0500  Min.  :-6.500000  Length:198430  
1st Qu.:0.1000 1st Qu.:-0.290000  Class :character  
Median :0.2000  Median : 0.000000  Mode  :character  
Mean   :0.1752  Mean   : 0.003367  
3rd Qu.:0.2500 3rd Qu.: 0.290000  
Max.   :0.3000  Max.   : 6.640000  
  
mf  
Length:198430  
Class :character  
Mode  :character
```

In RStudio, there is an additional function, `View()` (note the capital “V”) that opens the first 1000 rows (default) in the RStudio window, akin to a spreadsheet view.

```
View(ydat)
```

8. Data Frames

8.6. Accessing variables (columns) and subsetting

In R, `data.frames` can be subset similarly to other two-dimensional data structures. The `[` in R is used to denote subsetting of any kind. When working with two-dimensional data, we need two values inside the `[]` to specify the details. The specification is `[rows, columns]`. For example, to get the first three rows of `ydat`, use:

```
ydat[1:3, ]
```

```
symbol systematic_name nutrient rate expression
1   SFB2           YNL049C  Glucose  0.05    -0.24
2   <NA>           YNL095C  Glucose  0.05     0.28
3   QRI7           YDL104C  Glucose  0.05    -0.02
                                bp
1       ER to Golgi transport
2   biological process unknown
3 proteolysis and peptidolysis
                                mf
1   molecular function unknown
2   molecular function unknown
3 metalloendopeptidase activity
```

Note how the second number, the columns, is blank. R takes that to mean “all the columns”. Similarly, we can combine rows and columns specification arbitrarily.

```
ydat[1:3, 1:3]
```

```
symbol systematic_name nutrient
1   SFB2           YNL049C  Glucose
2   <NA>           YNL095C  Glucose
3   QRI7           YDL104C  Glucose
```

Because selecting a single variable, or column, is such a common operation, there are two shortcuts for doing so *with data.frames*. The first, the `$` operator works like so:

8. Data Frames

```
# Look at the column names, just to refresh memory  
colnames(ydat)  
  
[1] "symbol"           "systematic_name" "nutrient"  
[4] "rate"              "expression"      "bp"  
[7] "mf"  
  
# Note that I am using "head" here to limit the output  
head(ydat$symbol)
```

```
[1] "SFB2" NA      "QRI7" "CFT2" "SS02" "PSP2"
```

```
# What is the actual length of "symbol"?  
length(ydat$symbol)
```

```
[1] 198430
```

The second is related to the fact that, in R, data.frames are also lists. We subset a list by using `[[]]` notation. To get the second column of `ydat`, we can use:

```
head(ydat[[2]])
```

```
[1] "YNL049C" "YNL095C" "YDL104C" "YLR115W" "YMR183C"  
[6] "YML017W"
```

Alternatively, we can use the column name:

```
head(ydat[["systematic_name"]])
```

```
[1] "YNL049C" "YNL095C" "YDL104C" "YLR115W" "YMR183C"  
[6] "YML017W"
```

8.6.1. Some data exploration

There are a couple of columns that include numeric values. Which columns are numeric?

8. Data Frames

```
class(ydat$symbol)
```

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

```
class(ydat$rate)
```

```
[1] "numeric"
```

```
class(ydat$expression)
```

```
[1] "numeric"
```

Make histograms of: - the expression values - the rate values

What does the `table()` function do? Could you use that to look at the `rate` column given that that column appears to have repeated values?

What `rate` corresponds to the most nutrient-starved condition?

8.6.2. More advanced indexing and subsetting

We can use, for example, logical values (TRUE/FALSE) to subset data.frames.

```
head(ydat[ydat$symbol == 'LEU1', ])
```

	symbol	systematic_name	nutrient	rate	expression	bp
NA	<NA>	<NA>	<NA>	NA	NA	<NA>
NA.1	<NA>	<NA>	<NA>	NA	NA	<NA>
NA.2	<NA>	<NA>	<NA>	NA	NA	<NA>
NA.3	<NA>	<NA>	<NA>	NA	NA	<NA>
NA.4	<NA>	<NA>	<NA>	NA	NA	<NA>
NA.5	<NA>	<NA>	<NA>	NA	NA	<NA>
	mf					
NA	<NA>					

8. Data Frames

```
NA.1 <NA>
NA.2 <NA>
NA.3 <NA>
NA.4 <NA>
NA.5 <NA>
```

```
tail(ydat[ydat$symbol == 'LEU1', ])
```

```
symbol systematic_name nutrient rate expression
NA.47244 <NA> <NA> <NA> NA NA
NA.47245 <NA> <NA> <NA> NA NA
NA.47246 <NA> <NA> <NA> NA NA
NA.47247 <NA> <NA> <NA> NA NA
NA.47248 <NA> <NA> <NA> NA NA
NA.47249 <NA> <NA> <NA> NA NA
bp mf
NA.47244 <NA> <NA>
NA.47245 <NA> <NA>
NA.47246 <NA> <NA>
NA.47247 <NA> <NA>
NA.47248 <NA> <NA>
NA.47249 <NA> <NA>
```

What is the problem with this approach? It appears that there are a bunch of NA values. Taking a quick look at the `symbol` column, we see what the problem.

```
summary(ydat$symbol)
```

```
Length     Class      Mode
198430 character character
```

Using the `is.na()` function, we can make filter further to get down to values of interest.

```
head(ydat[ydat$symbol == 'LEU1' & !is.na(ydat$symbol), ])
```

8. Data Frames

```
symbol systematic_name nutrient rate expression
1526 LEU1 YGL009C Glucose 0.05 -1.12
7043 LEU1 YGL009C Glucose 0.10 -0.77
12555 LEU1 YGL009C Glucose 0.15 -0.67
18071 LEU1 YGL009C Glucose 0.20 -0.59
23603 LEU1 YGL009C Glucose 0.25 -0.20
29136 LEU1 YGL009C Glucose 0.30 0.03
                                bp
1526 leucine biosynthesis
7043 leucine biosynthesis
12555 leucine biosynthesis
18071 leucine biosynthesis
23603 leucine biosynthesis
29136 leucine biosynthesis
                                mf
1526 3-isopropylmalate dehydratase activity
7043 3-isopropylmalate dehydratase activity
12555 3-isopropylmalate dehydratase activity
18071 3-isopropylmalate dehydratase activity
23603 3-isopropylmalate dehydratase activity
29136 3-isopropylmalate dehydratase activity
```

Sometimes, looking at the data themselves is not that important. Using `dim()` is one possibility to look at the number of rows and columns after subsetting.

```
dim(ydat[ydat$expression > 3, ])
```

```
[1] 714 7
```

Find the high expressed genes when leucine-starved. For this task we can also use `subset` which allows us to treat column names as R variables (no \$ needed).

```
subset(ydat, nutrient == 'Leucine' & rate == 0.05 & expression > 3)
```

```
symbol systematic_name nutrient rate expression
133768 QDR2 YIL121W Leucine 0.05 4.61
133772 LEU1 YGL009C Leucine 0.05 3.84
```

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133858	BAP3	YDR046C	Leucine 0.05	4.29
135186	<NA>	YPL033C	Leucine 0.05	3.43
135187	<NA>	YLR267W	Leucine 0.05	3.23
135288	HXT3	YDR345C	Leucine 0.05	5.16
135963	TP02	YGR138C	Leucine 0.05	3.75
135965	YR02	YBR054W	Leucine 0.05	4.40
136102	GPG1	YGL121C	Leucine 0.05	3.08
136109	HSP42	YDR171W	Leucine 0.05	3.07
136119	HXT5	YHR096C	Leucine 0.05	4.90
136151	<NA>	YJL144W	Leucine 0.05	3.06
136152	MOH1	YBL049W	Leucine 0.05	3.43
136153	<NA>	YBL048W	Leucine 0.05	3.95
136189	HSP26	YBR072W	Leucine 0.05	4.86
136231	NCA3	YJL116C	Leucine 0.05	4.03
136233	<NA>	YBR116C	Leucine 0.05	3.28
136486	<NA>	YGR043C	Leucine 0.05	3.07
137443	ADH2	YMR303C	Leucine 0.05	4.15
137448	ICL1	YER065C	Leucine 0.05	3.54
137451	SFC1	YJR095W	Leucine 0.05	3.72
137569	MLS1	YNL117W	Leucine 0.05	3.76
			bp	
133768			multidrug transport	
133772			leucine biosynthesis	
133858			amino acid transport	
135186			meiosis*	
135187			biological process unknown	
135288			hexose transport	
135963			polyamine transport	
135965			biological process unknown	
136102			signal transduction	
136109			response to stress*	
136119			hexose transport	
136151			response to dessication	
136152			biological process unknown	
136153			<NA>	
136189			response to stress*	
136231	mitochondrion organization and biogenesis			
136233			<NA>	
136486			biological process unknown	
137443			fermentation*	
137448			glyoxylate cycle	

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137451	fumarate transport*
137569	glyoxylate cycle
	mf
133768	multidrug efflux pump activity
133772	3-isopropylmalate dehydratase activity
133858	amino acid transporter activity
135186	molecular function unknown
135187	molecular function unknown
135288	glucose transporter activity*
135963	spermine transporter activity
135965	molecular function unknown
136102	signal transducer activity
136109	unfolded protein binding
136119	glucose transporter activity*
136151	molecular function unknown
136152	molecular function unknown
136153	<NA>
136189	unfolded protein binding
136231	molecular function unknown
136233	<NA>
136486	transaldolase activity
137443	alcohol dehydrogenase activity
137448	isocitrate lyase activity
137451	succinate:fumarate antiporter activity
137569	malate synthase activity

8.7. Aggregating data

Aggregating data, or summarizing by category, is a common way to look for trends or differences in measurements between categories. Use `aggregate` to find the mean expression by gene symbol.

```
head(aggregate(ydat$expression, by=list( ydat$symbol), mean))
```

Group.1	x
1 AAC1	0.52888889
2 AAC3	-0.21628571
3 AAD10	0.43833333

8. Data Frames

```
4   AAD14 -0.07166667
5   AAD16  0.24194444
6   AAD4  -0.79166667

# or
head(aggregate(expression ~ symbol, mean, data=ydat))

symbol  expression
1   AAC1  0.52888889
2   AAC3 -0.21628571
3   AAD10 0.43833333
4   AAD14 -0.07166667
5   AAD16  0.24194444
6   AAD4  -0.79166667
```

8.8. Creating a data.frame from scratch

Sometimes it is useful to combine related data into one object. For example, let's simulate some data.

```
smoker = factor(rep(c("smoker", "non-smoker"), each=50))
smoker_numeric = as.numeric(smoker)
x = rnorm(100)
risk = x + 2*smoker_numeric
```

We have two variables, `risk` and `smoker` that are related. We can make a data.frame out of them:

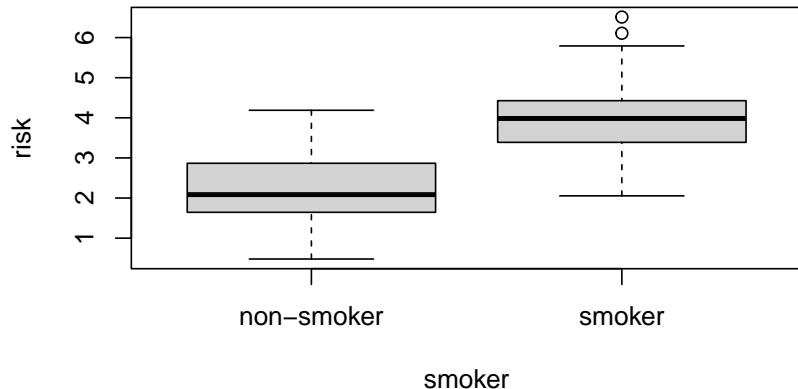
```
smoker_risk = data.frame(smoker = smoker, risk = risk)
head(smoker_risk)
```

```
smoker      risk
1 smoker 4.047227
2 smoker 3.710827
3 smoker 3.100671
4 smoker 4.497024
5 smoker 2.723650
6 smoker 2.860481
```

8. Data Frames

R also has plotting shortcuts that work with data.frames to simplify plotting

```
plot(risk ~ smoker, data=smoker_risk)
```



8.9. Saving a data.frame

Once we have a data.frame of interest, we may want to save it. The most portable way to save a data.frame is to use one of the `write` functions. In this case, let's save the data as a `.csv` file.

```
write.csv(smoker_risk, "smoker_risk.csv")
```

9. Factors

9.1. Factors

A factor is a special type of vector, normally used to hold a categorical variable—such as smoker/non-smoker, state of residency, zipcode—in many statistical functions. Such vectors have class “factor”. Factors are primarily used in Analysis of Variance (ANOVA) or other situations when “categories” are needed. When a factor is used as a predictor variable, the corresponding indicator variables are created (more later).

Note of caution that factors in R often *appear* to be character vectors when printed, but you will notice that they do not have double quotes around them. They are stored in R as numbers with a key name, so sometimes you will note that the factor *behaves* like a numeric vector.

```
# create the character vector
citizen<-c("uk","us","no","au","uk","us","us","no","au")

# convert to factor
citizenf<-factor(citizen)
citizen

[1] "uk" "us" "no" "au" "uk" "us" "us" "no" "au"

citizenf

[1] uk us no au uk us us no au
Levels: au no uk us
```

9. Factors

```
# convert factor back to character vector  
as.character(citizenf)
```

```
[1] "uk" "us" "no" "au" "uk" "us" "us" "no" "au"
```

```
# convert to numeric vector  
as.numeric(citizenf)
```

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

R stores many data structures as vectors with “attributes” and “class” (just so you have seen this).

```
attributes(citizenf)
```

```
$levels  
[1] "au" "no" "uk" "us"
```

```
$class  
[1] "factor"
```

```
class(citizenf)
```

```
[1] "factor"
```

```
# note that after unclassing, we can see the  
# underlying numeric structure again  
unclass(citizenf)
```

```
[1] 3 4 2 1 3 4 4 2 1  
attr(,"levels")  
[1] "au" "no" "uk" "us"
```

Tabulating factors is a useful way to get a sense of the “sample” set available.

9. Factors

```
table(citizenf)
```

```
citizenf  
au no uk us  
2 2 2 3
```

Part III.

Exploratory data analysis

Imagine you're on an adventure, about to embark on a journey into the unknown. You've just been handed a treasure map, with the promise of valuable insights waiting to be discovered. This map is your data set, and the journey is exploratory data analysis (EDA).

As you begin your exploration, you start by getting a feel for the terrain. You take a broad, bird's-eye view of the data, examining its structure and dimensions. Are you dealing with a vast landscape or a small, confined area? Are there any missing pieces in the map that you'll need to account for? Understanding the overall context of your data set is crucial before venturing further.

With a sense of the landscape, you now zoom in to identify key landmarks in the data. You might look for unusual patterns, trends, or relationships between variables. As you spot these landmarks, you start asking questions: What's causing that spike in values? Are these two factors related, or is it just a coincidence? By asking these questions, you're actively engaging with the data and forming hypotheses that could guide future analysis or experiments.

As you continue your journey, you realize that the map alone isn't enough to fully understand the terrain. You need more tools to bring the data to life. You start visualizing the data using charts, plots, and graphs. These visualizations act as your binoculars, allowing you to see patterns and relationships more clearly. Through them, you can uncover the hidden treasures buried within the data.

EDA isn't a linear path from start to finish. As you explore, you'll find yourself circling back to previous points, refining your questions, and digging deeper. The process is iterative, with each new discovery informing the next. And as you go, you'll gain a deeper understanding of the data's underlying structure and potential.

Finally, after your thorough exploration, you'll have a solid foundation to build upon. You'll be better equipped to make informed decisions, test hypotheses, and draw meaningful conclusions. The insights you've gained through EDA will serve as a compass, guiding you towards the true value hidden within

your data. And with that, you've successfully completed your journey through exploratory data analysis.

10. Introduction to `dplyr`: mammal sleep dataset

The dataset we will be using to introduce the `dplyr` package is an updated and expanded version of the mammals sleep dataset. Updated sleep times and weights were taken from V. M. Savage and G. B. West. A quantitative, theoretical framework for understanding mammalian sleep¹.

10.1. Learning goals

- Know that `dplyr` is just a different approach to manipulating data in `data.frames`.
- List the commonly used `dplyr` verbs and how they can be used to manipulate `data.frames`.
- Show how to aggregate and summarize data using `dplyr`
- Know what the piping operator, `|>`, is and how it can be used.

10.2. Learning objectives

- Select subsets of the mammal sleep dataset.
- Reorder the dataset.
- Add columns to the dataset based on existing columns.
- Summarize the amount of sleep by categorical variables using `group_by` and `summarize`.

¹A quantitative, theoretical framework for understanding mammalian sleep. Van M. Savage, Geoffrey B. West. Proceedings of the National Academy of Sciences Jan 2007, 104 (3) 1051-1056; DOI: [10.1073/pnas.0610080104](https://doi.org/10.1073/pnas.0610080104)

10. Introduction to dplyr: mammal sleep dataset

10.3. What is dplyr?

The *dplyr* package is a specialized package for working with `data.frames` (and the related `tibble`) to transform and summarize tabular data with rows and columns. For another explanation of *dplyr* see the *dplyr* package vignette: [Introduction to dplyr](#)

10.4. Why Is dplyr useful?

dplyr contains a set of functions—commonly called the *dplyr* “verbs”—that perform common data manipulations such as filtering for rows, selecting specific columns, re-ordering rows, adding new columns and summarizing data. In addition, *dplyr* contains a useful function to perform another common task which is the “split-apply-combine” concept.

Compared to base functions in R, the functions in *dplyr* are often easier to work with, are more consistent in the syntax and are targeted for data analysis around data frames, instead of just vectors.

10.5. Data: Mammals Sleep

The `msleep` (mammals sleep) data set contains the sleep times and weights for a set of mammals and is available in the `dagdata` repository on github. This data set contains 83 rows and 11 variables. The data happen to be available as a `dataset` in the `ggplot2` package. To get access to the `msleep` dataset, we need to first install the `ggplot2` package.

```
install.packages('ggplot2')
```

Then, we can load the library.

```
library(ggplot2)
data(msleep)
```

10. Introduction to dplyr: mammal sleep dataset

As with many datasets in R, “help” is available to describe the dataset itself.

```
?msleep
```

The columns are described in the help page, but are included here, also.

column name	Description
name	common name
genus	taxonomic rank
vore	carnivore, omnivore or herbivore?
order	taxonomic rank
conservation	the conservation status of the mammal
sleep_total	total amount of sleep, in hours
sleep_rem	rem sleep, in hours
sleep_cycle	length of sleep cycle, in hours
awake	amount of time spent awake, in hours
brainwt	brain weight in kilograms
bodywt	body weight in kilograms

10.6. dplyr verbs

The dplyr verbs are listed here. There are many other functions available in dplyr, but we will focus on just these.

dplyr verbs	Description
<code>select()</code>	select columns
<code>filter()</code>	filter rows
<code>arrange()</code>	re-order or arrange rows
<code>mutate()</code>	create new columns
<code>summarise()</code>	summarise values
<code>group_by()</code>	allows for group operations in the “split-apply-combine” concept

10.7. Using the dplyr verbs

The two most basic functions are `select()` and `filter()`, which selects columns and filters rows respectively. What are the equivalent ways to select columns without dplyr? And filtering to include only specific rows?

Before proceeding, we need to install the dplyr package:

```
install.packages('dplyr')
```

And then load the library:

```
library(dplyr)
```

```
Attaching package: 'dplyr'
```

```
The following objects are masked from 'package:stats':
```

```
filter, lag
```

```
The following objects are masked from 'package:base':
```

```
intersect, setdiff, setequal, union
```

10.7.1. Selecting columns: `select()`

Select a set of columns such as the `name` and the `sleep_total` columns.

```
sleepData <- select(msleep, name, sleep_total)
head(sleepData)
```

```
# A tibble: 6 x 2
  name          sleep_total
  <chr>           <dbl>
1 Cheetah        12.1
```

10. Introduction to dplyr: mammal sleep dataset

```
2 Owl monkey           17
3 Mountain beaver      14.4
4 Greater short-tailed shrew 14.9
5 Cow                  4
6 Three-toed sloth     14.4
```

To select all the columns *except* a specific column, use the “-” (subtraction) operator (also known as negative indexing). For example, to select all columns except `name`:

```
head(select(msleep, -name))
```

```
# A tibble: 6 x 10
  genus      vore   order  conservation sleep_total sleep_rem sleep_cycle awake
  <chr>      <chr>  <chr>    <chr>          <dbl>      <dbl>      <dbl>      <dbl>
1 Acinonyx   carni  Carnivo~ lc            12.1       NA        NA       11.9
2 Aotus       omni   Primates <NA>          17         1.8       NA        7
3 Aplodontia herbi  Rodentia nt            14.4       2.4       NA       9.6
4 Blarina     omni   Soricom~ lc            14.9       2.3       0.133    9.1
5 Bos         herbi  Artioda~ domesticated  4          0.7       0.667    20
6 Bradypus    herbi  Pilosa   <NA>          14.4       2.2       0.767    9.6
# i 2 more variables: brainwt <dbl>, bodywt <dbl>
```

To select a range of columns by name, use the “`:`” operator. Note that dplyr allows us to use the column names without quotes and as “indices” of the columns.

```
head(select(msleep, name:order))
```

```
# A tibble: 6 x 4
  name              genus      vore   order
  <chr>             <chr>    <chr>  <chr>
1 Cheetah           Acinonyx   carni  Carnivora
2 Owl monkey        Aotus      omni   Primates
3 Mountain beaver   Aplodontia herbi  Rodentia
4 Greater short-tailed shrew Blarina   omni   Soricomorpha
5 Cow                Bos        herbi  Artiodactyla
6 Three-toed sloth Bradypus    herbi  Pilosa
```

10. Introduction to dplyr: mammal sleep dataset

To select all columns that start with the character string “sl”, use the function `starts_with()`.

```
head(select(msleep, starts_with("sl")))
```

```
# A tibble: 6 x 3
  sleep_total sleep_rem sleep_cycle
    <dbl>      <dbl>      <dbl>
1     12.1        NA        NA
2      17         1.8        NA
3     14.4         2.4        NA
4     14.9         2.3     0.133
5       4          0.7      0.667
6     14.4         2.2     0.767
```

Some additional options to select columns based on a specific criteria include:

1. `ends_with()` = Select columns that end with a character string
2. `contains()` = Select columns that contain a character string
3. `matches()` = Select columns that match a regular expression
4. `one_of()` = Select column names that are from a group of names

10.7.2. Selecting rows: `filter()`

The `filter()` function allows us to filter rows to include only those rows that *match* the filter. For example, we can filter the rows for mammals that sleep a total of more than 16 hours.

```
filter(msleep, sleep_total >= 16)
```

```
# A tibble: 8 x 11
  name   genus vore  order conservation sleep_total sleep_rem sleep_cycle awake
  <chr>  <chr> <chr> <chr> <chr>      <dbl>      <dbl>      <dbl> <dbl>
1 Owl    mo~ Aotus omni Prim~ <NA>           17        1.8       NA       7
```

10. Introduction to dplyr: mammal sleep dataset

```

2 Long-n~ Dasy~ carni Cing~ lc          17.4      3.1      0.383    6.6
3 North ~ Dide~ omni  Dide~ lc          18        4.9      0.333     6
4 Big br~ Epte~ inse~ Chir~ lc          19.7      3.9      0.117    4.3
5 Thick-- Lutr~ carni Dide~ lc          19.4      6.6      NA       4.6
6 Little~ Myot~ inse~ Chir~ <NA>        19.9      2        0.2      4.1
7 Giant ~ Prio~ inse~ Cing~ en          18.1      6.1      NA       5.9
8 Arctic~ Sper~ herbi Rode~ lc          16.6      NA      NA       7.4
# i 2 more variables: brainwt <dbl>, bodywt <dbl>

```

Filter the rows for mammals that sleep a total of more than 16 hours *and* have a body weight of greater than 1 kilogram.

```
filter(msleep, sleep_total >= 16, bodywt >= 1)
```

```

# A tibble: 3 x 11
  name   genus vore  order conservation sleep_total sleep_rem sleep_cycle awake
  <chr>  <chr> <chr> <chr>           <dbl>      <dbl>      <dbl> <dbl>
1 Long-n~ Dasy~ carni Cing~ lc          17.4      3.1      0.383    6.6
2 North ~ Dide~ omni  Dide~ lc          18        4.9      0.333     6
3 Giant ~ Prio~ inse~ Cing~ en          18.1      6.1      NA       5.9
# i 2 more variables: brainwt <dbl>, bodywt <dbl>

```

Filter the rows for mammals in the Perissodactyla and Primates taxonomic order. The `%in%` operator is a logical operator that returns TRUE for values of a vector that are present *in* a second vector.

```
filter(msleep, order %in% c("Perissodactyla", "Primates"))
```

```

# A tibble: 15 x 11
  name   genus vore  order conservation sleep_total sleep_rem sleep_cycle awake
  <chr>  <chr> <chr> <chr>           <dbl>      <dbl>      <dbl> <dbl>
1 Owl m~ Aotus omni  Prim~ <NA>        17        1.8      NA       7
2 Grivet Cerc~ omni  Prim~ lc          10        0.7      NA      14
3 Horse  Equus herbi Peri~ domesticated 2.9        0.6      1      21.1
4 Donkey Equus herbi Peri~ domesticated 3.1        0.4      NA      20.9
5 Patas~ Eryt~ omni  Prim~ lc          10.9      1.1      NA      13.1
6 Galago Gala~ omni  Prim~ <NA>        9.8        1.1      0.55    14.2
7 Human  Homo  omni  Prim~ <NA>        8         1.9      1.5      16

```

10. Introduction to dplyr: mammal sleep dataset

```
8 Mongo~ Lemur herbi Prim~ vu           9.5    0.9    NA   14.5
 9 Macaq~ Maca~ omni  Prim~ <NA>        10.1   1.2    0.75  13.9
10 Slow ~ Nyct~ carni Prim~ <NA>       11     NA    NA    13
11 Chimp~ Pan   omni  Prim~ <NA>        9.7    1.4    1.42  14.3
12 Baboon Papio omni  Prim~ <NA>        9.4    1     0.667 14.6
13 Potto  Pero~ omni  Prim~ lc          11     NA    NA    13
14 Squir~ Saim~ omni  Prim~ <NA>        9.6    1.4    NA   14.4
15 Brazi~ Tapi~ herbi Peri~ vu          4.4    1     0.9   19.6
# i 2 more variables: brainwt <dbl>, bodywt <dbl>
```

You can use the boolean operators (e.g. `>`, `<`, `>=`, `<=`, `!=`, `%in%`) to create the logical tests.

10.8. “Piping” with `|>`

It is not unusual to want to perform a set of operations using dplyr. The pipe operator `|>` allows us to “pipe” the output from one function into the input of the next. While there is nothing special about how R treats operations that are written in a pipe, the idea of piping is to allow us to read multiple functions operating one after another from left-to-right. Without piping, one would either 1) save each step in set of functions as a temporary variable and then pass that variable along the chain or 2) have to “nest” functions, which can be hard to read.

Here’s an example we have already used:

```
head(select(msleep, name, sleep_total))
```

```
# A tibble: 6 x 2
  name                 sleep_total
  <chr>                <dbl>
1 Cheetah              12.1
2 Owl monkey            17
3 Mountain beaver      14.4
4 Greater short-tailed shrew 14.9
5 Cow                  4
6 Three-toed sloth    14.4
```

10. Introduction to dplyr: mammal sleep dataset

Now in this case, we will pipe the `msleep` data frame to the function that will select two columns (`name` and `sleep_total`) and then pipe the new data frame to the function `head()`, which will return the head of the new data frame.

```
msleep |>
  select(name, sleep_total) |>
  head()
```

```
# A tibble: 6 x 2
  name           sleep_total
  <chr>          <dbl>
1 Cheetah        12.1
2 Owl monkey     17
3 Mountain beaver 14.4
4 Greater short-tailed shrew 14.9
5 Cow             4
6 Three-toed sloth 14.4
```

You will soon see how useful the pipe operator is when we start to combine many functions.

Now that you know about the pipe operator (`|>`), we will use it throughout the rest of this tutorial.

10.8.1. Arrange Or Re-order Rows Using `arrange()`

To arrange (or re-order) rows by a particular column, such as the taxonomic order, list the name of the column you want to arrange the rows by:

```
msleep |> arrange(order) |> head()
```

```
# A tibble: 6 x 11
  name   genus vore  order conservation sleep_total sleep_rem sleep_cycle awake
  <chr>  <chr> <chr> <chr> <chr>          <dbl>      <dbl>      <dbl> <dbl>
1 Tenrec  Tenr~ omni  Afro~ <NA>            15.6       2.3       NA    8.4
2 Cow     Bos    herbi Arti~ domesticated     4         0.7      0.667  20
3 Roe de~ Capr~ herbi Arti~ lc              3         NA       NA    21
```

10. Introduction to dplyr: mammal sleep dataset

```
4 Goat      Capri herbi Arti~ lc          5.3      0.6      NA    18.7
5 Giraffe   Gira~ herbi Arti~ cd          1.9      0.4      NA    22.1
6 Sheep     Ovis  herbi Arti~ domesticated 3.8      0.6      NA    20.2
# i 2 more variables: brainwt <dbl>, bodywt <dbl>
```

Now we will select three columns from msleep, arrange the rows by the taxonomic order and then arrange the rows by sleep_total. Finally, show the head of the final data frame:

```
msleep |>
  select(name, order, sleep_total) |>
  arrange(order, sleep_total) |>
  head()
```

```
# A tibble: 6 x 3
  name      order      sleep_total
  <chr>     <chr>     <dbl>
1 Tenrec    Afrosoricida 15.6
2 Giraffe   Artiodactyla  1.9
3 Roe deer  Artiodactyla  3
4 Sheep     Artiodactyla  3.8
5 Cow       Artiodactyla  4
6 Goat      Artiodactyla  5.3
```

Same as above, except here we filter the rows for mammals that sleep for 16 or more hours, instead of showing the head of the final data frame:

```
msleep |>
  select(name, order, sleep_total) |>
  arrange(order, sleep_total) |>
  filter(sleep_total >= 16)
```

```
# A tibble: 8 x 3
  name      order      sleep_total
  <chr>     <chr>     <dbl>
1 Big brown bat Chiroptera 19.7
2 Little brown bat Chiroptera 19.9
3 Long-nosed armadillo Cingulata 17.4
```

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```
4 Giant armadillo      Cingulata          18.1
5 North American Opossum Didelphimorphia   18
6 Thick-tailed oposum  Didelphimorphia    19.4
7 Owl monkey           Primates           17
8 Arctic ground squirrel Rodentia         16.6
```

For something slightly more complicated do the same as above, except arrange the rows in the sleep_total column in a descending order. For this, use the function `desc()`

```
msleep |>
  select(name, order, sleep_total) |>
  arrange(order, desc(sleep_total)) |>
  filter(sleep_total >= 16)
```

```
# A tibble: 8 x 3
  name            order      sleep_total
  <chr>           <chr>        <dbl>
1 Little brown bat Chiroptera     19.9
2 Big brown bat   Chiroptera     19.7
3 Giant armadillo Cingulata      18.1
4 Long-nosed armadillo Cingulata     17.4
5 Thick-tailed oposum Didelphimorphia 19.4
6 North American Opossum Didelphimorphia 18
7 Owl monkey       Primates       17
8 Arctic ground squirrel Rodentia     16.6
```

10.9. Create New Columns Using `mutate()`

The `mutate()` function will add new columns to the data frame. Create a new column called `rem_proportion`, which is the ratio of rem sleep to total amount of sleep.

```
msleep |>
  mutate(rem_proportion = sleep_rem / sleep_total) |>
  head()
```

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```
# A tibble: 6 x 12
  name   genus vore  order conservation sleep_total sleep_rem sleep_cycle awake
  <chr>  <chr> <chr> <chr>           <dbl>      <dbl>      <dbl> <dbl>
1 Cheetah Acin~ carni Carn~ lc        12.1       NA       NA    11.9
2 Owl     mo~ Aotus omni Prim~ <NA>      17        1.8      NA     7
3 Mounta~ Aplo~ herbi Rode~ nt       14.4       2.4      NA    9.6
4 Greate~ Blar~ omni Sori~ lc       14.9       2.3     0.133   9.1
5 Cow      Bos   herbi Arti~ domesticated 4        0.7     0.667   20
6 Three~~ Brad~ herbi Pilo~ <NA>      14.4       2.2     0.767   9.6
# i 3 more variables: brainwt <dbl>, bodywt <dbl>, rem_proportion <dbl>
```

You can add many new columns using `mutate` (separated by commas). Here we add a second column called `bodywt_grams` which is the `bodywt` column in grams.

```
msleep |>
  mutate(rem_proportion = sleep_rem / sleep_total,
        bodywt_grams = bodywt * 1000) |>
  head()
```

```
# A tibble: 6 x 13
  name   genus vore  order conservation sleep_total sleep_rem sleep_cycle awake
  <chr>  <chr> <chr> <chr>           <dbl>      <dbl>      <dbl> <dbl>
1 Cheetah Acin~ carni Carn~ lc        12.1       NA       NA    11.9
2 Owl     mo~ Aotus omni Prim~ <NA>      17        1.8      NA     7
3 Mounta~ Aplo~ herbi Rode~ nt       14.4       2.4      NA    9.6
4 Greate~ Blar~ omni Sori~ lc       14.9       2.3     0.133   9.1
5 Cow      Bos   herbi Arti~ domesticated 4        0.7     0.667   20
6 Three~~ Brad~ herbi Pilo~ <NA>      14.4       2.2     0.767   9.6
# i 4 more variables: brainwt <dbl>, bodywt <dbl>, rem_proportion <dbl>,
#   bodywt_grams <dbl>
```

Is there a relationship between `rem_proportion` and `bodywt`?
How about `sleep_total`?

10.9.1. Create summaries: `summarise()`

The `summarise()` function will create summary statistics for a given column in the data frame such as finding the mean.

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For example, to compute the average number of hours of sleep, apply the `mean()` function to the column `sleep_total` and call the summary value `avg_sleep`.

```
msleep |>
  summarise(avg_sleep = mean(sleep_total))
```

```
# A tibble: 1 x 1
  avg_sleep
  <dbl>
1     10.4
```

There are many other summary statistics you could consider such `sd()`, `min()`, `max()`, `median()`, `sum()`, `n()` (returns the length of vector), `first()` (returns first value in vector), `last()` (returns last value in vector) and `n_distinct()` (number of distinct values in vector).

```
msleep |>
  summarise(avg_sleep = mean(sleep_total),
            min_sleep = min(sleep_total),
            max_sleep = max(sleep_total),
            total = n())
```

```
# A tibble: 1 x 4
  avg_sleep min_sleep max_sleep total
  <dbl>      <dbl>      <dbl> <int>
1     10.4       1.9      19.9     83
```

10.10. Grouping data: `group_by()`

The `group_by()` verb is an important function in dplyr. The `group_by` allows us to use the concept of “split-apply-combine”. We literally want to split the data frame by some variable (e.g. taxonomic order), apply a function to the individual data frames and then combine the output. This approach is similar to the `aggregate` function from R, but `group_by` integrates with dplyr.

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Let's do that: split the msleep data frame by the taxonomic order, then ask for the same summary statistics as above. We expect a set of summary statistics for each taxonomic order.

```
msleep |>
  group_by(order) |>
  summarise(avg_sleep = mean(sleep_total),
            min_sleep = min(sleep_total),
            max_sleep = max(sleep_total),
            total = n())
```

```
# A tibble: 19 x 5
  order      avg_sleep min_sleep max_sleep total
  <chr>        <dbl>     <dbl>     <dbl> <int>
1 Afrosoricida    15.6      15.6      15.6    1
2 Artiodactyla     4.52       1.9       9.1     6
3 Carnivora       10.1       3.5      15.8    12
4 Cetacea          4.5        2.7       5.6     3
5 Chiroptera      19.8      19.7      19.9    2
6 Cingulata        17.8      17.4      18.1    2
7 Didelphimorphia   18.7       18       19.4    2
8 Diprotodontia    12.4      11.1      13.7    2
9 Erinaceomorpha    10.2      10.1      10.3    2
10 Hyracoidea      5.67       5.3       6.3     3
11 Lagomorpha       8.4        8.4       8.4     1
12 Monotremata     8.6        8.6       8.6     1
13 Perissodactyla   3.47       2.9       4.4     3
14 Pilosa           14.4      14.4      14.4    1
15 Primates          10.5       8        17     12
16 Proboscidea       3.6        3.3       3.9     2
17 Rodentia          12.5       7        16.6    22
18 Scandentia        8.9        8.9       8.9     1
19 Soricomorpha     11.1       8.4      14.9     5
```

11. Case Study: Behavioral Risk Factor Surveillance System

11.1. A Case Study on the Behavioral Risk Factor Surveillance System

The Behavioral Risk Factor Surveillance System (BRFSS) is a large-scale health survey conducted annually by the Centers for Disease Control and Prevention (CDC) in the United States. The BRFSS collects information on various health-related behaviors, chronic health conditions, and the use of preventive services among the adult population (18 years and older) through telephone interviews. The main goal of the BRFSS is to identify and monitor the prevalence of risk factors associated with chronic diseases, inform public health policies, and evaluate the effectiveness of health promotion and disease prevention programs. The data collected through BRFSS is crucial for understanding the health status and needs of the population, and it serves as a valuable resource for researchers, policy makers, and healthcare professionals in making informed decisions and designing targeted interventions.

In this chapter, we will walk through an exploratory data analysis (EDA) of the Behavioral Risk Factor Surveillance System dataset using R. EDA is an important step in the data analysis process, as it helps you to understand your data, identify trends, and detect any anomalies before performing more advanced analyses. We will use various R functions and packages to explore the dataset, with a focus on active learning and hands-on experience.

11.2. Loading the Dataset

First, let's load the dataset into R. We will use the `read.csv()` function from the base R package to read the data and store it in a data frame called `brfss`. Make sure the CSV file is in your working directory, or provide the full path to the file.

First, we need to get the data. Either download the data from [THIS LINK](#) or have R do it directly from the command-line (preferred):

```
download.file('https://raw.githubusercontent.com/seandavi/ITR/master/BRFSS-subset.csv',
              destfile = 'BRFSS-subset.csv')

path <- file.choose()      # look for BRFSS-subset.csv

stopifnot(file.exists(path))
brfss <- read.csv(path)
```

11.3. Inspecting the Data

Once the data is loaded, let's take a look at the first few rows of the dataset using the `head()` function:

```
head(brfss)
```

	Age	Weight	Sex	Height	Year
1	31	48.98798	Female	157.48	1990
2	57	81.64663	Female	157.48	1990
3	43	80.28585	Male	177.80	1990
4	72	70.30682	Male	170.18	1990
5	31	49.89516	Female	154.94	1990
6	58	54.43108	Female	154.94	1990

This will display the first six rows of the dataset, allowing you to get a feel for the data structure and variable types.

Next, let's check the dimensions of the dataset using the `dim()` function:

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```
dim(brfss)
```

```
[1] 20000      5
```

This will return the number of rows and columns in the dataset, which is important to know for subsequent analyses.

11.4. Summary Statistics

Now that we have a basic understanding of the data structure, let's calculate some summary statistics. The `summary()` function in R provides a quick overview of the main statistics for each variable in the dataset:

```
summary(brfss)
```

Age	Weight	Sex	Height
Min. :18.00	Min. : 34.93	Length:20000	Min. :105.0
1st Qu.:36.00	1st Qu.: 61.69	Class :character	1st Qu.:162.6
Median :51.00	Median : 72.57	Mode :character	Median :168.0
Mean :50.99	Mean : 75.42		Mean :169.2
3rd Qu.:65.00	3rd Qu.: 86.18		3rd Qu.:177.8
Max. :99.00	Max. :278.96		Max. :218.0
NA's :139	NA's :649		NA's :184

Year
Min. :1990
1st Qu.:1990
Median :2000
Mean :2000
3rd Qu.:2010
Max. :2010

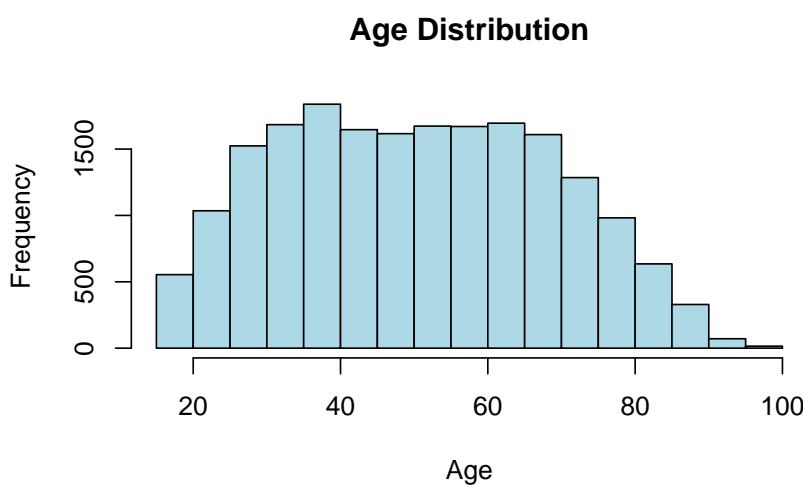
This will display the minimum, first quartile, median, mean, third quartile, and maximum for each numeric variable, and the frequency counts for each factor level for categorical variables.

11.5. Data Visualization

Visualizing the data can help you identify patterns and trends in the dataset. Let's start by creating a histogram of the Age variable using the `hist()` function.

This will create a histogram showing the frequency distribution of ages in the dataset. You can customize the appearance of the histogram by adjusting the parameters within the `hist()` function.

```
hist(brfss$Age, main = "Age Distribution",
      xlab = "Age", col = "lightblue")
```



💡 What are the options for a histogram?

The `hist()` function has many options. For example, you can change the number of bins, the color of the bars, the title, and the x-axis label. You can also add a vertical line at the mean or median, or add a normal curve to the histogram. For more information, type `?hist` in the R console.

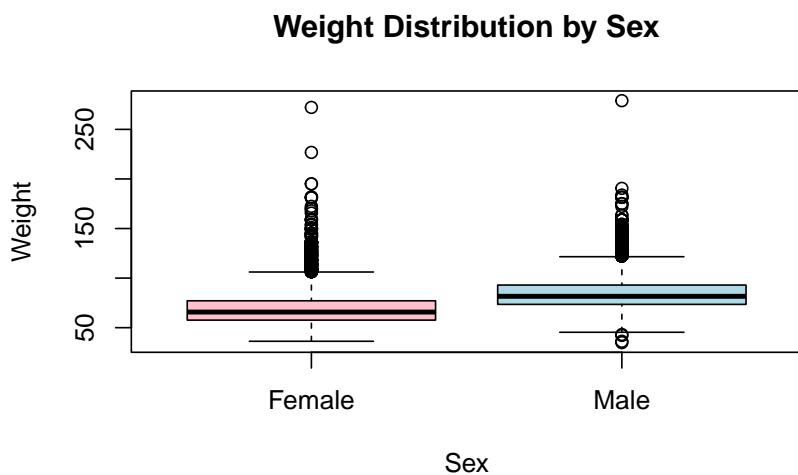
More generally, it is important to understand the options available for each function you use. You can do this by reading the documentation for the function, which can be accessed by typing `?function_name` or

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`help("function_name")` in the R console.

Next, let's create a boxplot to compare the distribution of Weight between males and females. We will use the `boxplot()` function for this. This will create a boxplot comparing the weight distribution between males and females. You can customize the appearance of the boxplot by adjusting the parameters within the `boxplot()` function.

```
boxplot(brfss$Weight ~ brfss$Sex, main = "Weight Distribution by Sex",
       xlab = "Sex", ylab = "Weight", col = c("pink", "lightblue"))
```



11.6. Analyzing Relationships Between Variables

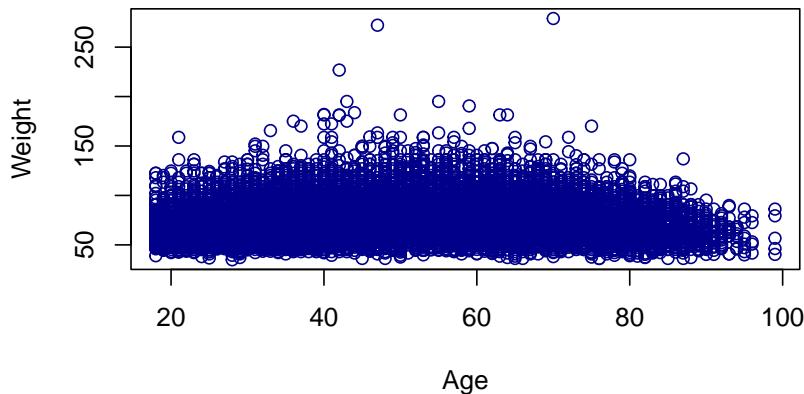
To further explore the data, let's investigate the relationship between age and weight using a scatterplot. We will use the `plot()` function for this:

This will create a scatterplot of age and weight, allowing you to visually assess the relationship between these two variables.

```
plot(brfss$Age, brfss$Weight, main = "Scatterplot of Age and Weight",
      xlab = "Age", ylab = "Weight", col = "darkblue")
```

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Scatterplot of Age and Weight



To quantify the strength of the relationship between age and weight, we can calculate the correlation coefficient using the `cor()` function:

This will return the correlation coefficient between age and weight, which can help you determine whether there is a linear relationship between these variables.

```
cor(brfss$Age, brfss$Weight)
```

```
[1] NA
```

Why does `cor()` give a value of `NA`? What can we do about it? A quick glance at `help("cor")` will give you the answer.

```
cor(brfss$Age, brfss$Weight, use = "complete.obs")
```

```
[1] 0.02699989
```

11.7. Exercises

1. What is the mean weight in this dataset? How about the median? What is the difference between the two? What does this tell you about the distribution of weights in the dataset?

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```
mean(brfss$Weight, na.rm = TRUE)
```

```
[1] 75.42455
```

```
median(brfss$Weight, na.rm = TRUE)
```

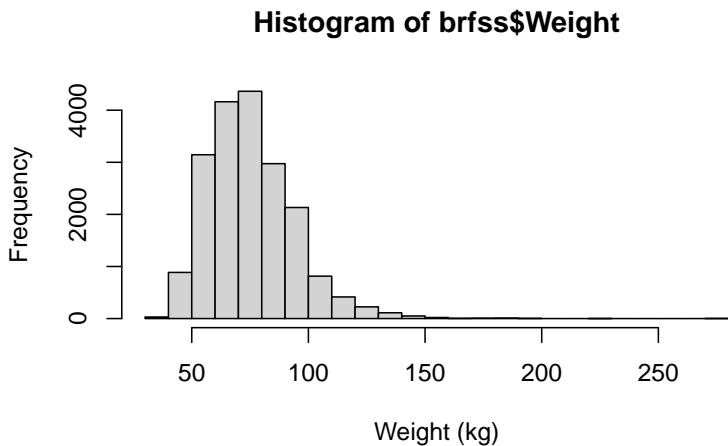
```
[1] 72.57478
```

```
mean(brfss$Weight, na.rm=TRUE) - median(brfss$Weight, na.rm = TRUE)
```

```
[1] 2.849774
```

- Given the findings about the `mean` and `median` in the previous exercise, use the `hist()` function to create a histogram of the weight distribution in this dataset. How would you describe the shape of this distribution?

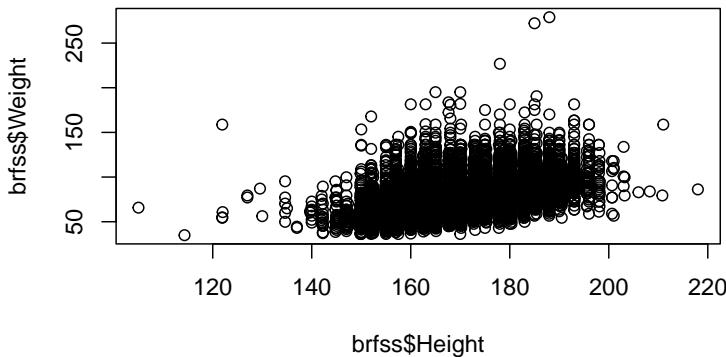
```
hist(brfss$Weight, xlab="Weight (kg)", breaks = 30)
```



- Use `plot()` to examine the relationship between height and weight in this dataset.

```
plot(brfss$Height, brfss$Weight)
```

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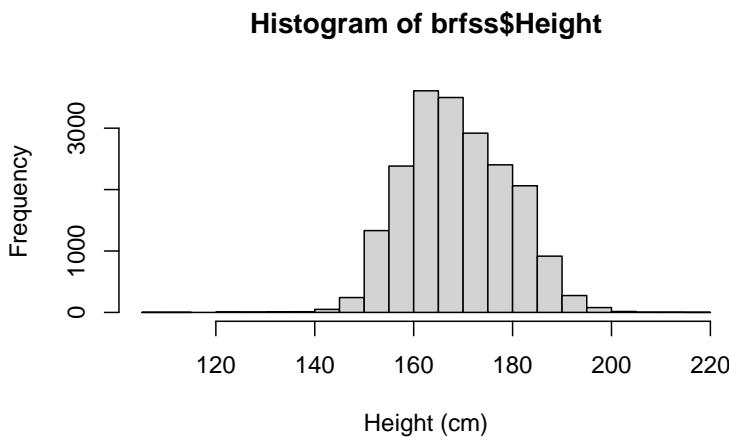
4. What is the correlation between height and weight? What does this tell you about the relationship between these two variables?

```
cor(brfss$Height, brfss$Weight, use = "complete.obs")
```

[1] 0.5140928

5. Create a histogram of the height distribution in this dataset. How would you describe the shape of this distribution?

```
hist(brfss$Height, xlab="Height (cm)", breaks = 30)
```



11.8. Conclusion

In this chapter, we have demonstrated how to perform an exploratory data analysis on the Behavioral Risk Factor Surveil-

11. Case Study: Behavioral Risk Factor Surveillance System

lance System dataset using R. We covered data loading, inspection, summary statistics, visualization, and the analysis of relationships between variables. By actively engaging with the R code and data, you have gained valuable experience in using R for EDA and are well-equipped to tackle more complex analyses in your future work.

Remember that EDA is just the beginning of the data analysis process, and further statistical modeling and hypothesis testing will likely be necessary to draw meaningful conclusions from your data. However, EDA is a crucial step in understanding your data and informing your subsequent analyses.

11.9. Learn about the data

Using the data exploration techniques you have seen to explore the brfss dataset.

- `summary()`
- `dim()`
- `colnames()`
- `head()`
- `tail()`
- `class()`
- `View()`

You may want to investigate individual columns visually using plotting like `hist()`. For categorical data, consider using something like `table()`.

11.10. Clean data

R read Year as an integer value, but it's really a factor

```
brfss$Year <- factor(brfss$Year)
```

11. Case Study: Behavioral Risk Factor Surveillance System

11.11. Weight in 1990 vs. 2010 Females

- Create a subset of the data

```
brfssFemale <- brfss[brfss$Sex == "Female",]  
summary(brfssFemale)
```

Age	Weight	Sex	Height
Min. :18.00	Min. : 36.29	Length:12039	Min. :105.0
1st Qu.:37.00	1st Qu.: 57.61	Class :character	1st Qu.:157.5
Median :52.00	Median : 65.77	Mode :character	Median :163.0
Mean :51.92	Mean : 69.05		Mean :163.3
3rd Qu.:67.00	3rd Qu.: 77.11		3rd Qu.:168.0
Max. :99.00	Max. :272.16		Max. :200.7
NA's :103	NA's :560		NA's :140

Year
1990:5718
2010:6321

- Visualize

```
plot(Weight ~ Year, brfssFemale)
```



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- Statistical test

```
t.test(Weight ~ Year, brfssFemale)
```

```
Welch Two Sample t-test

data: Weight by Year
t = -27.133, df = 11079, p-value < 2.2e-16
alternative hypothesis: true difference in means between group 1990 and group 2010 is not equal to zero
95 percent confidence interval:
-8.723607 -7.548102
sample estimates:
mean in group 1990 mean in group 2010
64.81838      72.95424
```

11.12. Weight and height in 2010 Males

- Create a subset of the data

```
brfss2010Male <- subset(brfss, Year == 2010 & Sex == "Male")
summary(brfss2010Male)
```

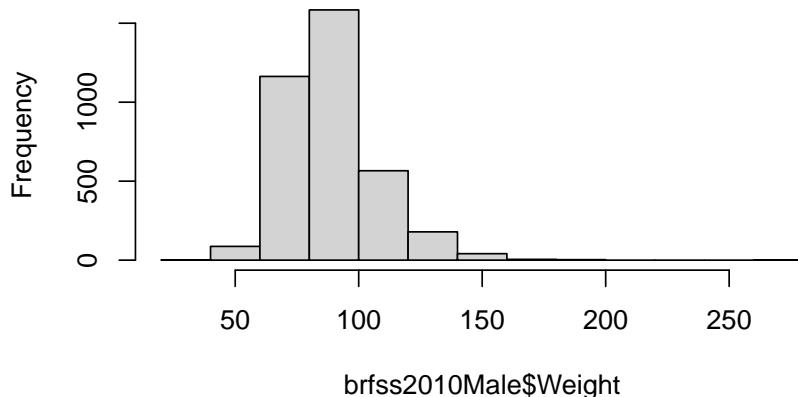
Age	Weight	Sex	Height	Year
Min. :18.00	Min. : 36.29	Length:3679	Min. :135	1990: 0
1st Qu.:45.00	1st Qu.: 77.11	Class :character	1st Qu.:173	2010:3679
Median :57.00	Median : 86.18	Mode :character	Median :178	
Mean :56.25	Mean : 88.85		Mean :178	
3rd Qu.:68.00	3rd Qu.: 99.79		3rd Qu.:183	
Max. :99.00	Max. :278.96		Max. :218	
NA's :30	NA's :49		NA's :31	

- Visualize the relationship

```
hist(brfss2010Male$Weight)
```

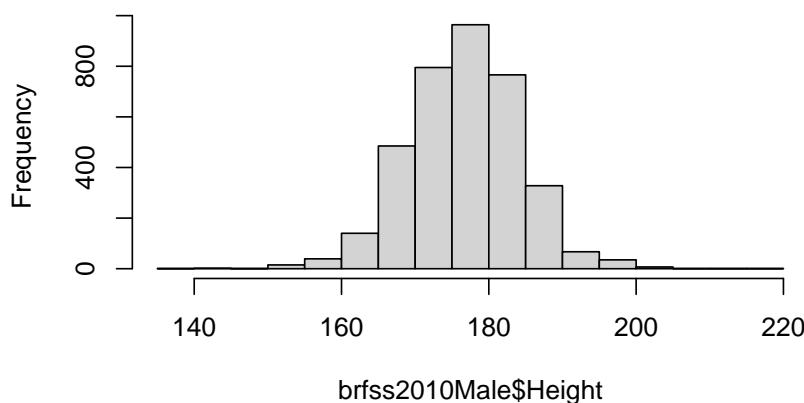
11. Case Study: Behavioral Risk Factor Surveillance System

Histogram of brfss2010Male\$Weight



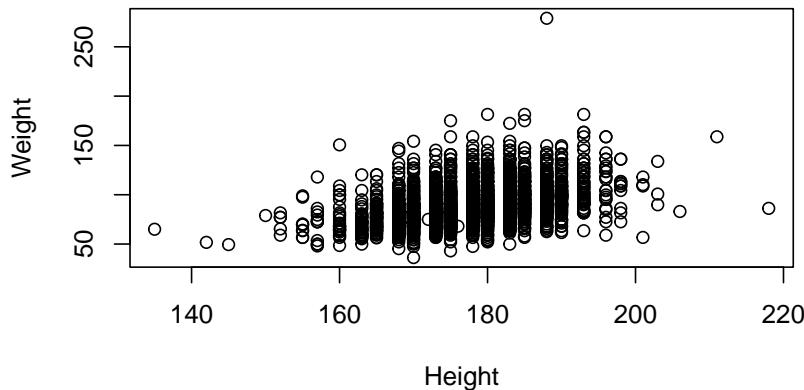
```
hist(brfss2010Male$Height)
```

Histogram of brfss2010Male\$Height



```
plot(Weight ~ Height, brfss2010Male)
```

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- Fit a linear model (regression)

```
fit <- lm(Weight ~ Height, brfss2010Male)  
fit
```

```
Call:  
lm(formula = Weight ~ Height, data = brfss2010Male)
```

```
Coefficients:  
(Intercept) Height  
-86.8747 0.9873
```

Summarize as ANOVA table

```
anova(fit)
```

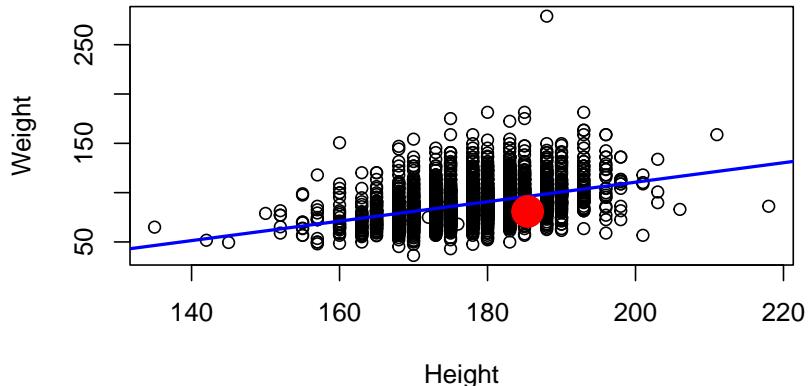
Analysis of Variance Table

```
Response: Weight  
          Df Sum Sq Mean Sq F value    Pr(>F)  
Height      1 197664 197664   693.8 < 2.2e-16 ***  
Residuals 3617 1030484      285  
---  
Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

- Plot points, superpose fitted regression line; where am I?

11. Case Study: Behavioral Risk Factor Surveillance System

```
plot(Weight ~ Height, brfss2010Male)
abline(fit, col="blue", lwd=2)
# Substitute your own weight and height...
points(73 * 2.54, 178 / 2.2, col="red", cex=4, pch=20)
```



- Class and available ‘methods’

```
class(fit)                  # 'noun'
methods(class=class(fit))   # 'verb'
```

- Diagnostics

```
plot(fit)
# Note that the "plot" above does not have a ".lm"
# However, R will use "plot.lm". Why?
?plot.lm
```

Part IV.

statistics

12. Working with distribution functions

Which values do pnorm, dnorm, qnorm, and rnorm return?
How do I remember the difference between these?

I find it helpful to have visual representations of distributions as pictures. It is difficult for me to think of distributions, or differences between probability, density, and quantiles without visualizing the shape of the distribution. So I figured it would be helpful to have a visual guide to pnorm, dnorm, qnorm, and rnorm.

Table 12.1.: Table 1.1: Functions for the normal distribution

Function	Input	Output
pnorm	x	$P(X < x)$
dnorm	x	$f(x)$, or the height of the density curve at x
qnorm	q, a quantile from 0 to 1	x such that $P(X < x) = q$
rnorm	n	n random samples from the distribution

12.1. pnorm

This function gives the probability function for a normal distribution. If you do not specify the mean and standard deviation, R defaults to standard normal. Figure 12.1

```
pnorm(q, mean = 0, sd = 1, lower.tail = TRUE, log.p = FALSE)
```

The R help file for pnorm provides the template above. The value you input for q is a value on the x-axis, and the returned value is the area under the distribution curve to the left of that point.

12. Working with distribution functions

```
Warning: Using `size` aesthetic for lines was deprecated in ggplot2 3.4.0.  
i Please use `linewidth` instead.
```

This function gives the probability function for a normal distribution. If you do not specify the mean and standard deviation, R defaults to standard normal.

`pnorm(q, mean = 0, sd = 1, lower.tail = TRUE, log.p = FALSE)` The R help file for pnorm provides the template above. The value you input for q is a value on the x-axis, and the returned value is the area under the distribution curve to the left of that point.

The option `lower.tail = TRUE` tells R to use the area to the left of the given point. This is the default, so will remain true even without entering it. In order to compute the area to the right of the given point, you can either switch to `lower.tail = FALSE`, or simply calculate `1-pnorm()` instead. This is demonstrated below.

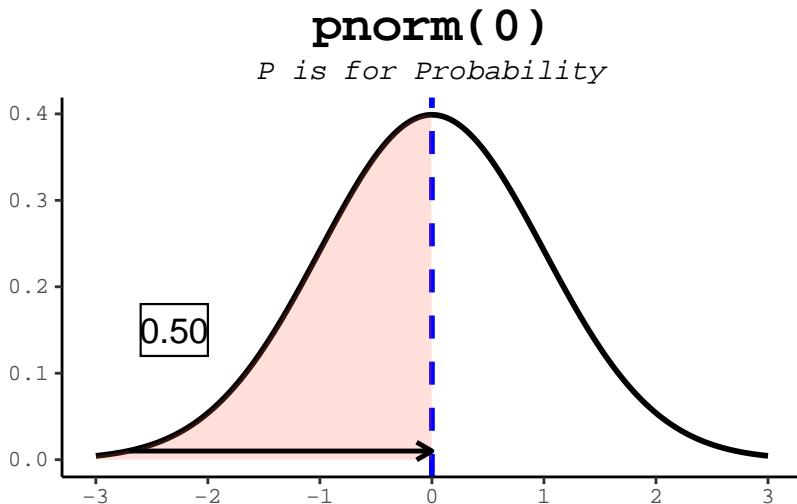


Figure 12.1.: The pnorm function takes a quantile (value on the x-axis) and returns the area under the curve to the left of that value.

The option `lower.tail = TRUE` tells R to use the area to the left of the given point. This is the default, so will remain true even without entering it. In order to compute the area to the right of the given point, you can either switch to `lower.tail = FALSE`, or simply calculate `1-pnorm()` instead.

12. Working with distribution functions

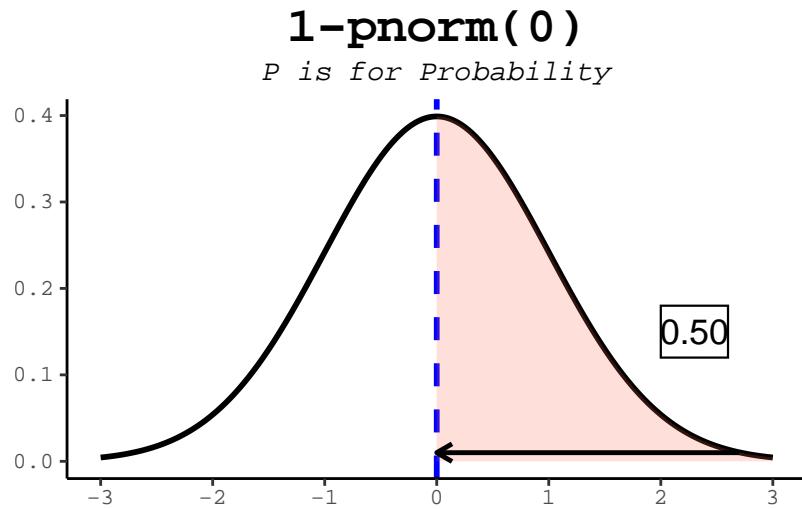


Figure 12.2.: The pnorm function takes a quantile (value on the x-axis) and returns the area under the curve to the left of that value.

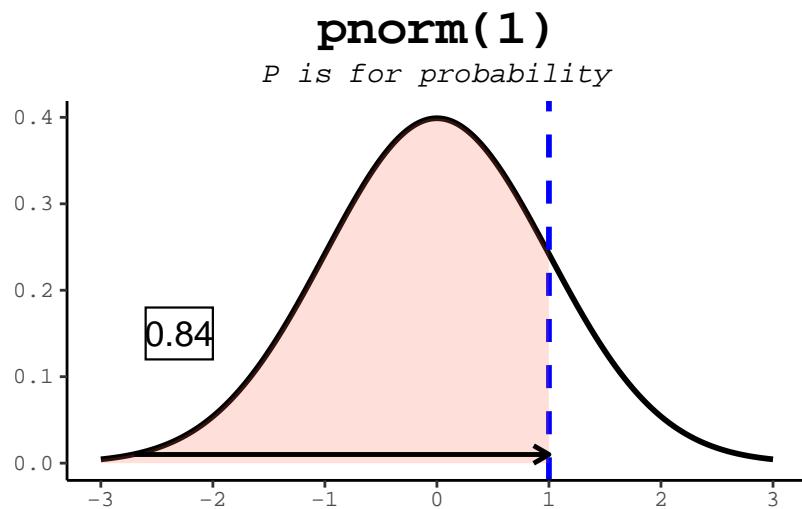


Figure 12.3.: The pnorm function takes a quantile (value on the x-axis) and returns the area under the curve to the left of that value.

12. Working with distribution functions

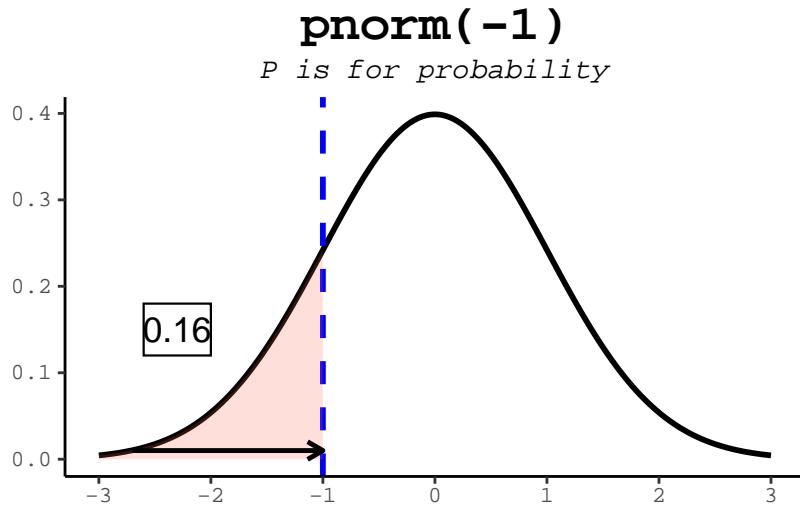


Figure 12.4.: The `pnorm` function takes a quantile (value on the x-axis) and returns the area under the curve to the left of that value.

12.2. dnorm

This function calculates the probability density function (PDF) for the normal distribution. It gives the probability density (height of the curve) at a specified value (x).

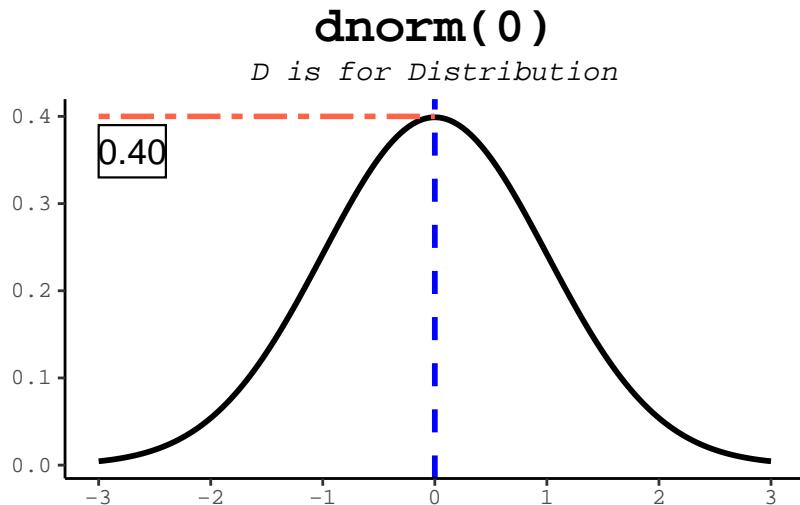


Figure 12.5.: The `dnorm` function returns the height of the normal distribution at a given point.

12.3. qnorm

This function calculates the quantiles of the normal distribution. It returns the value (x) corresponding to a specified probability

12. Working with distribution functions

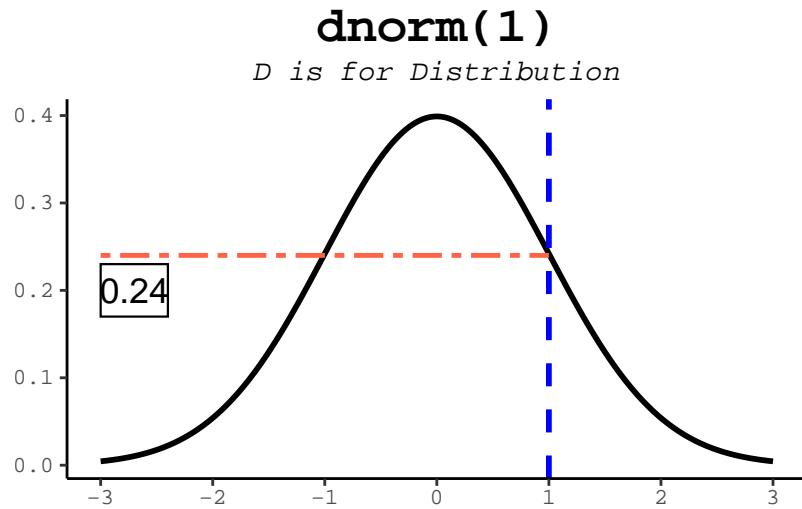


Figure 12.6.: The `dnorm` function returns the height of the normal distribution at a given point.

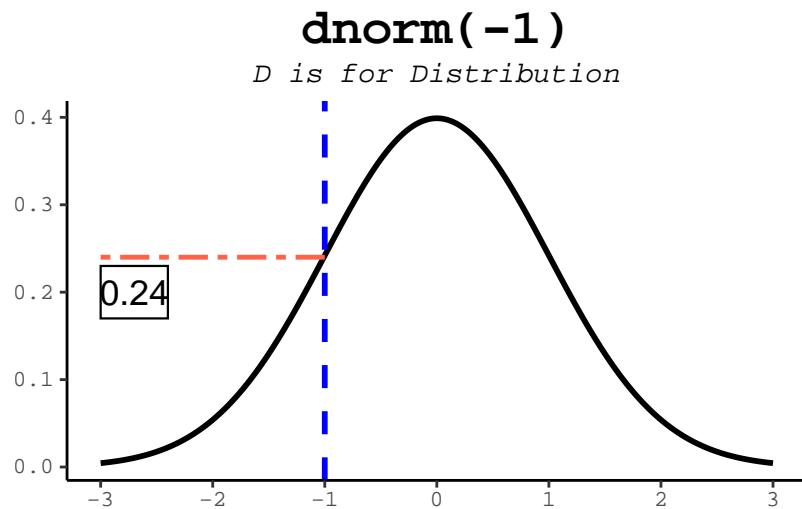


Figure 12.7.: The `dnorm` function returns the height of the normal distribution at a given point.

12. Working with distribution functions

(p). It is the inverse of the `pnorm` function.

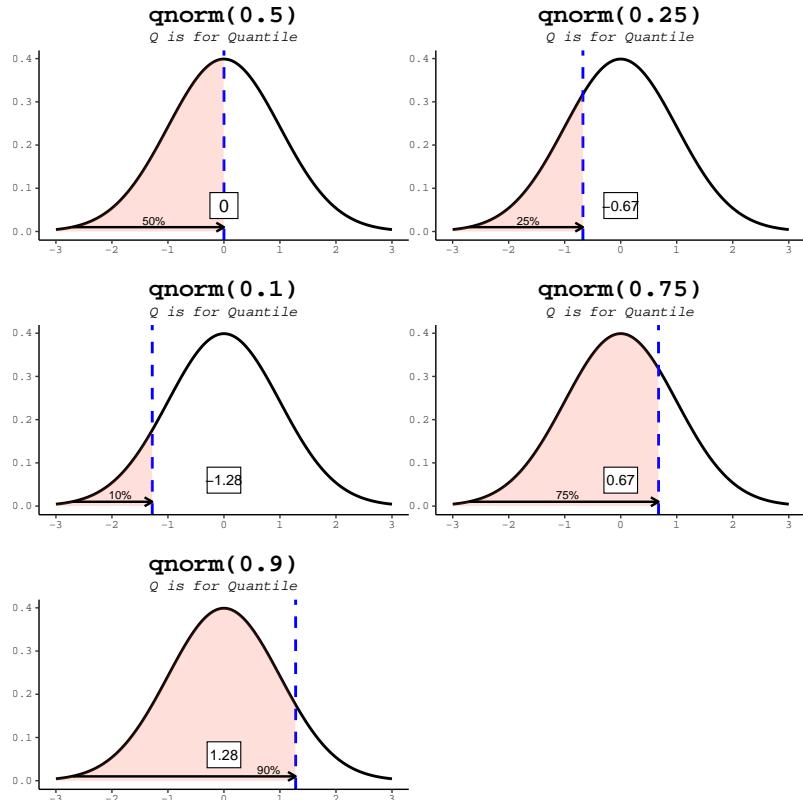


Figure 12.9.: The `qnorm` function is the inverse of the `pnorm` function in that it takes a probability and gives the quantile.

Figure 12.10.: The `qnorm` function is the inverse of the `pnorm` function in that it takes a probability and gives the quantile.

Figure 12.12.: The `qnorm` function is the inverse of the `pnorm` function in that it takes a probability and gives the quantile.

12.4. `rnorm`

```
print(r1)
```

12. Working with distribution functions

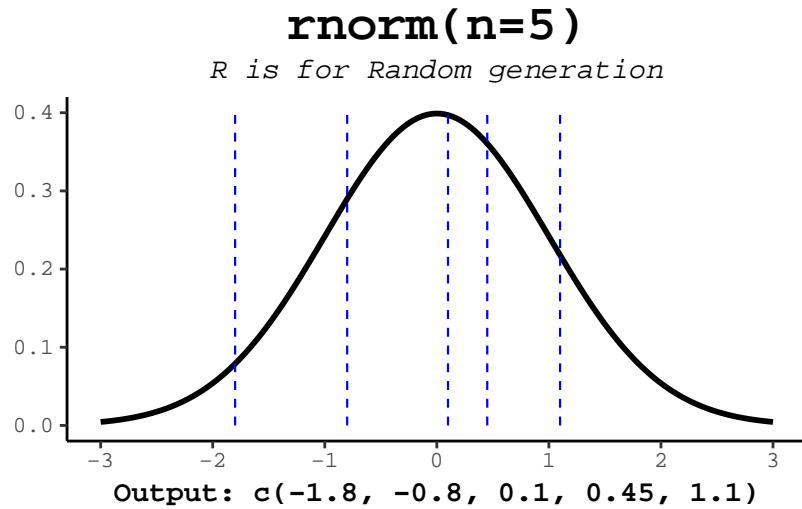


Figure 12.13.: The `rnorm` function takes a number of samples and returns a vector of random numbers from the normal distribution (with `mean=0, sd=1` as defaults)

12.5. IQ scores

Normal Distribution and its Application with IQ

The normal distribution, also known as the Gaussian distribution, is a continuous probability distribution characterized by its bell-shaped curve. It is defined by two parameters: the mean (μ) and the standard deviation (σ). The mean represents the central tendency of the distribution, while the standard deviation represents the dispersion or spread of the data.

The IQ scores are an excellent example of the normal distribution, as they are designed to follow this distribution pattern. The mean IQ score is set at 100, and the standard deviation is set at 15. This means that the majority of the population (about 68%) have an IQ score between 85 and 115, while 95% of the population have an IQ score between 70 and 130.

- What is the probability of having an IQ score between 85 and 115?

```
pnorm(115, mean = 100, sd = 15) - pnorm(85, mean = 100, sd = 15)
```

- What is the 90th percentile of the IQ scores?

```
qnorm(0.9, mean = 100, sd = 15)
```

12. Working with distribution functions

- What is the probability of having an IQ score above 130?

```
1 - pnorm(130, mean = 100, sd = 15)
```

- What is the probability of having an IQ score below 70?

```
pnorm(70, mean = 100, sd = 15)
```

13. The t-statistic and t-distribution

13.1. Background

The t-test is a [statistical hypothesis test](#) that is commonly used when the data are normally distributed (follow a normal distribution) if the value of the population standard deviation were known. When the population standard deviation is not known and is replaced by an estimate based on the data, the test statistic follows a Student's t distribution.

T-tests are handy hypothesis tests in statistics when you want to compare means. You can compare a sample mean to a hypothesized or target value using a one-sample t-test. You can compare the means of two groups with a two-sample t-test. If you have two groups with paired observations (e.g., before and after measurements), use the paired t-test.

A t-test looks at the t-statistic, the t-distribution values, and the degrees of freedom to determine the statistical significance. To conduct a test with three or more means, we would use an analysis of variance.

The distribution that the t-statistic follows was described in a famous paper (Student 1908) by “Student”, a pseudonym for [William Sealy Gosset](#).

13.2. The Z-score and probability

Before talking about the t-distribution and t-scores, let's review the Z-score, its relation to the normal distribution, and probability.

13. The t -statistic and t -distribution

The Z-score is defined as:

$$Z = \frac{x - \mu}{\sigma} \quad (13.1)$$

where μ is the population mean from which x is drawn and σ is the population standard deviation (taken as known, not estimated from the data).

The probability of observing a Z score of z or greater can be calculated by $pnorm(z, \mu, \sigma)$.

For example, let's assume that our "population" is known and it truly has a mean 0 and standard deviation 1. If we have observations drawn from that population, we can assign a probability of seeing that observation by random chance *under the assumption that the null hypothesis is TRUE*.

```
zscores = seq(-5, 5, 1)
```

For each value of `zscores`, let's calculate the p-value and put the results in a `data.frame`.

```
df = data.frame(
  zscore = zscores,
  pval   = pnorm(zscore, 0, 1)
)
df
```

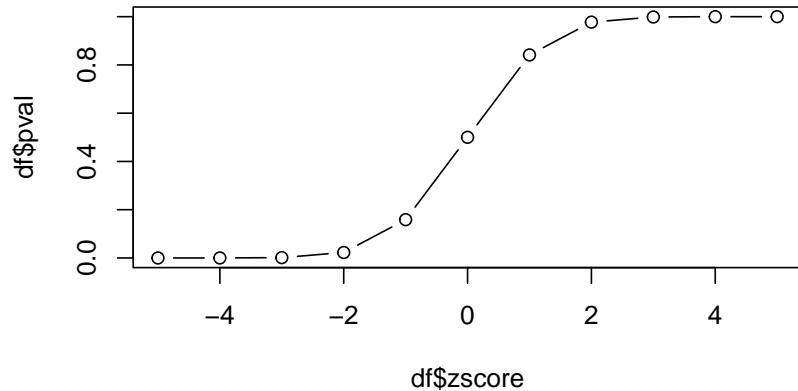

	zscore	pval
1	-5	2.866516e-07
2	-4	3.167124e-05
3	-3	1.349898e-03
4	-2	2.275013e-02
5	-1	1.586553e-01
6	0	5.000000e-01
7	1	8.413447e-01
8	2	9.772499e-01
9	3	9.986501e-01
10	4	9.999683e-01
11	5	9.999997e-01

13. The t -statistic and t -distribution

Why is the p-value of something 5 population standard deviations away from the mean ($zscore=5$) nearly 1 in this calculation? What is the default for `pnorm` with respect to being one-sided or two-sided?

Let's plot the values of probability vs z-score:

```
plot(df$zscore, df$pval, type='b')
```



This plot is the *empirical* cumulative density function (cdf) for our data. How can we use it? If we know the z-score, we can look up the probability of observing that value. Since we have constructed our experiment to follow the standard normal distribution, this cdf also represents the cdf of the standard normal distribution.

13.2.1. Small diversion: two-sided `pnorm` function

The `pnorm` function returns the “one-sided” probability of having a value at least as extreme as the observed x and uses the “lower” tail by default. Let’s create a function that computes two-sided p-values.

1. Take the absolute value of x
2. Compute `pnorm` with `lower.tail=FALSE` so we get lower p-values with larger values of x .
3. Since we want to include both tails, we need to multiply the area (probability) returned by `pnorm` by 2.

13. The *t*-statistic and *t*-distribution

```
twosidedpnorm = function(x, mu=0, sd=1) {  
  2*pnorm(abs(x), mu, sd, lower.tail=FALSE)  
}
```

And we can test this to see how likely it is to be 2 or 3 standard deviations from the mean:

```
twosidedpnorm(2)
```

```
[1] 0.04550026
```

```
twosidedpnorm(3)
```

```
[1] 0.002699796
```

13.3. The *t*-distribution

We spent time above working with z-scores and probability. An important aspect of working with the normal distribution is that we MUST assume that we know the standard deviation. Remember that the Z-score is defined as:

$$Z = \frac{x - \mu}{\sigma}$$

The formula for the *population* standard deviation is:

$$\sigma = \sqrt{\frac{1}{N} \sum_{i=1}^N (xi - \mu)^2} \quad (13.2)$$

In general, the population standard deviation is taken as “known” as we did above.

If we do not but only have a *sample* from the population, instead of using the Z-score, we use the t-score defined as:

$$t = \frac{x - \bar{x}}{s} \quad (13.3)$$

13. The *t*-statistic and *t*-distribution

This looks quite similar to the formula for Z-score, but here we have to *estimate* the standard deviation, s from the data. The formula for s is:

$$s = \sqrt{\frac{1}{N-1} \sum_{i=1}^N (x_i - \bar{x})^2} \quad (13.4)$$

Since we are estimating the standard deviation from the data, this leads to extra variability that shows up as “fatter tails” for smaller sample sizes than for larger sample sizes. We can see this by comparing the *t-distribution* for various numbers of degrees of freedom (sample sizes).

We can look at the effect of sample size on the distributions graphically by looking at the densities for 3, 5, 10, 20 degrees of freedom and the normal distribution:

```
library(dplyr)
library(ggplot2)
t_values = seq(-6,6,0.01)
df = data.frame(
  value = t_values,
  t_3    = dt(t_values,3),
  t_6    = dt(t_values,6),
  t_10   = dt(t_values,10),
  t_20   = dt(t_values,20),
  Normal= dnorm(t_values)
) |>
  tidyr::gather("Distribution", "density", -value)
ggplot(df, aes(x=value, y=density, color=Distribution)) +
  geom_line()
```

13. The t-statistic and t-distribution

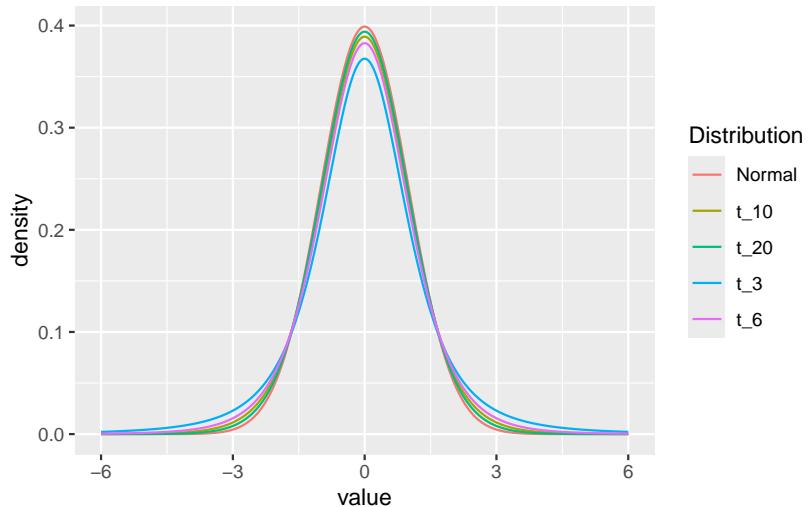
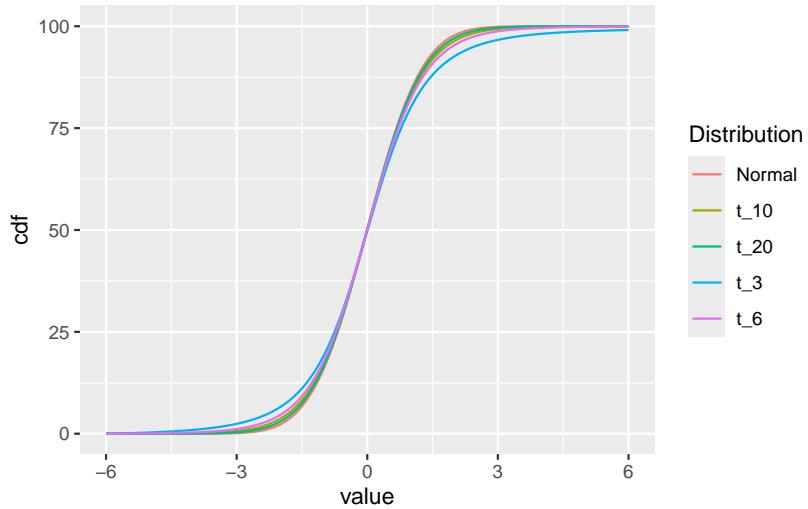


Figure 13.1.: t-distributions for various degrees of freedom. Note that the tails are fatter for smaller degrees of freedom, which is a result of estimating the standard deviation from the data.

The `dt` and `dnorm` functions give the density of the distributions for each point.

```
df2 = df |>
  group_by(Distribution) |>
  arrange(value) |>
  mutate(cdf=cumsum(density))
ggplot(df2, aes(x=value, y=cdf, color=Distribution)) +
  geom_line()
```



13. The *t*-statistic and *t*-distribution

13.3.1. p-values based on Z vs t

When we have a “sample” of data and want to compute the statistical significance of the difference of the mean from the population mean, we calculate the standard deviation of the sample means (standard error).

$$z = \frac{x - \mu}{\sigma / \sqrt{n}}$$

Let’s look at the relationship between the p-values of Z (from the normal distribution) vs t for a **sample** of data.

```
set.seed(5432)
samp = rnorm(5,mean = 0.5)
z = sqrt(length(samp)) * mean(samp) #simplifying assumption (sigma=1, mu=0)
```

And the p-value if we assume we know the standard deviation:

```
pnorm(z, lower.tail = FALSE)
```

```
[1] 0.02428316
```

In reality, we don’t know the standard deviation, so we have to estimate it from the data. We can do this by calculating the sample standard deviation:

```
ts = sqrt(length(samp)) * mean(samp) / sd(samp)
pnorm(ts, lower.tail = FALSE)
```

```
[1] 0.0167297
```

```
pt(ts,df = length(samp)-1, lower.tail = FALSE)
```

```
[1] 0.0503001
```

13. The t-statistic and t-distribution

13.3.2. Experiment

When sampling from a normal distribution, we often calculate p-values to test hypotheses or determine the statistical significance of our results. The p-value represents the probability of obtaining a test statistic as extreme or more extreme than the one observed, under the null hypothesis.

In a typical scenario, we assume that the population mean and standard deviation are known. However, in many real-life situations, we don't know the true population standard deviation, and we have to estimate it using the sample standard deviation (Equation 13.4). This estimation introduces some uncertainty into our calculations, which affects the p-values. When we include an estimate of the standard deviation, we switch from using the standard normal (z) distribution to the t-distribution for calculating p-values.

What would happen if we used the normal distribution to calculate p-values when we use the sample standard deviation? Let's find out!

1. Simulate a bunch of samples of size n from the standard normal distribution
2. Calculate the p-value distribution for those samples based on the normal.
3. Calculate the p-value distribution for those samples based on the normal, but with the *estimated* standard deviation.
4. Calculate the p-value distribution for those samples based on the t-distribution.

Create a function that draws a sample of size n from the standard normal distribution.

```
zf = function(n) {  
  samp = rnorm(n)  
  z = sqrt(length(samp)) * mean(samp) / 1 #simplifying assumption (sigma=1, mu=0)  
  z  
}
```

And give it a try:

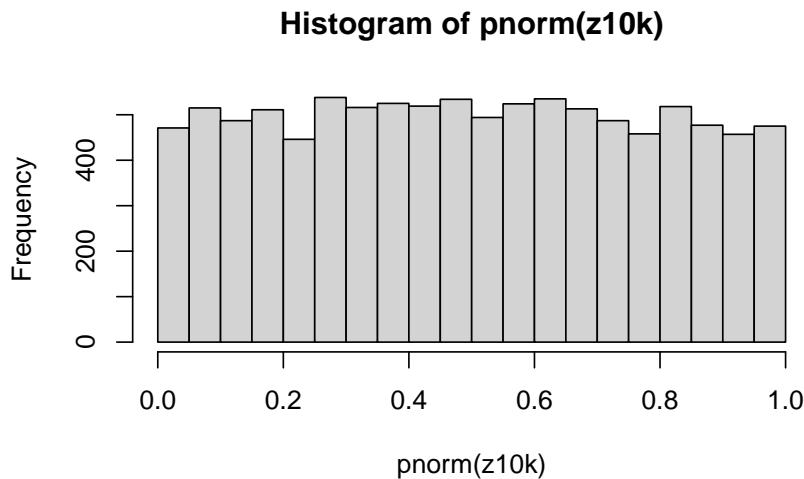
13. The t-statistic and t-distribution

```
zf(5)
```

```
[1] 0.7406094
```

Perform 10000 replicates of our sampling and z-scoring. We are using the assumption that we know the population standard deviation; in this case, we do know since we are sampling from the standard normal distribution.

```
z10k = replicate(10000,zf(5))
hist(pnorm(z10k))
```



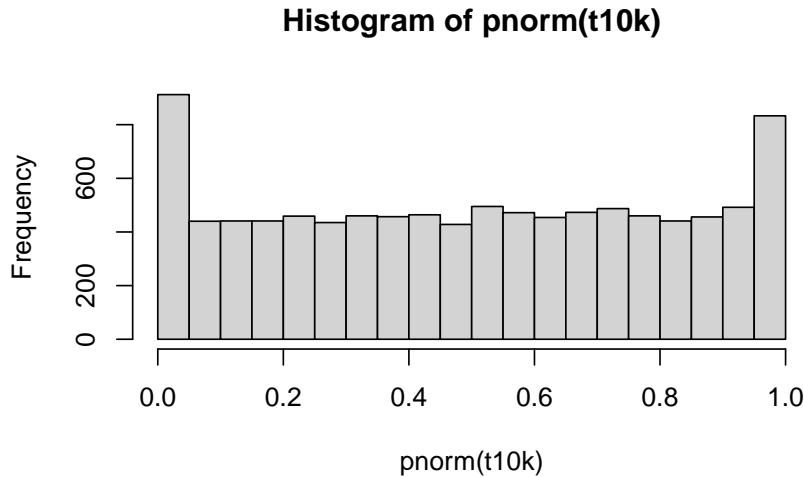
And do the same, but now creating a t-score function. We are using the assumption that we *don't* know the population standard deviation; in this case, we must estimate it from the data. Note the difference in the calculation of the t-score (`ts`) as compared to the z-score (`z`).

```
tf = function(n) {
  samp = rnorm(n)
  # now, using the sample standard deviation since we
  # "don't know" the population standard deviation
  ts = sqrt(length(samp)) * mean(samp) / sd(samp)
  ts
}
```

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If we use those t-scores and calculate the p-values based on the normal distribution, the histogram of those p-values looks like:

```
t10k = replicate(10000,tf(5))
hist(pnorm(t10k))
```

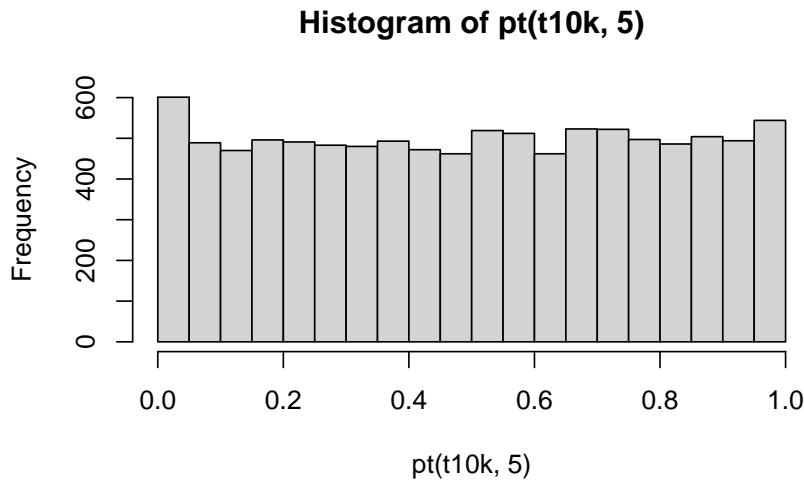


Since we are using the normal distribution to calculate the p-values, we are, in effect, assuming that we know the population standard deviation. This assumption is incorrect, and we can see that the p-values are not uniformly distributed between 0 and 1.

If we use those t-scores and calculate the p-values based on the t-distribution, the histogram of those p-values looks like:

13. The t -statistic and t -distribution

```
hist(pt(t10k, 5))
```

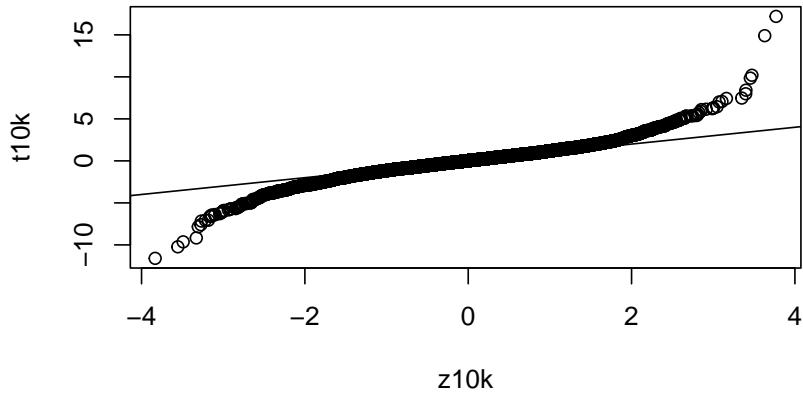


Now, the p-values are uniformly distributed between 0 and 1, as expected.

What is a qqplot and how do we use it? A qqplot is a plot of the quantiles of two distributions against each other. If the two distributions are identical, the points will fall on a straight line. If the two distributions are different, the points will deviate from the straight line. We can use a qqplot to compare the t -distribution to the normal distribution. If the t -distribution is identical to the normal distribution, the points will fall on a straight line. If the t -distribution is different from the normal distribution, the points will deviate from the straight line. In this case, we can see that the t -distribution is different from the normal distribution, as the points deviate from the straight line. What would happen if we increased the sample size? The t -distribution would approach the normal distribution, and the points would fall closer and closer to the straight line.

```
qqplot(z10k,t10k)
abline(0,1)
```

13. The *t*-statistic and *t*-distribution



13.4. Summary of *t*-distribution vs normal distribution

The *t*-distribution is a family of probability distributions that depends on a parameter called degrees of freedom, which is related to the sample size. The *t*-distribution approaches the standard normal distribution as the sample size increases but has heavier tails for smaller sample sizes. This means that the *t*-distribution is more conservative in calculating p-values for small samples, making it harder to reject the null hypothesis. Including an estimate of the standard deviation changes the way we calculate p-values by switching from the standard normal distribution to the *t*-distribution, which accounts for the uncertainty introduced by estimating the population standard deviation from the sample. This adjustment is particularly important for small sample sizes, as it provides a more accurate assessment of the statistical significance of our results.

13.5. `t.test`

13.5.1. One-sample

We are going to use the `t.test` function to perform a one-sample *t*-test. The `t.test` function takes a vector of values as input that represents the sample values. In this case, we'll

13. The *t*-statistic and *t*-distribution

simulate our sample using the `rnorm` function and presume that our “effect-size” is 1.

```
x = rnorm(20,1)
# small sample
# Just use the first 5 values of the sample
t.test(x[1:5])
```

One Sample t-test

```
data: x[1:5]
t = 0.97599, df = 4, p-value = 0.3843
alternative hypothesis: true mean is not equal to 0
95 percent confidence interval:
-1.029600 2.145843
sample estimates:
mean of x
0.5581214
```

In this case, we set up the experiment so that the null hypothesis is true (the true mean is not zero, but actually 1). However, we only have a small sample size that leads to a modest p-value.

Increasing the sample size allows us to see the effect more clearly.

```
t.test(x[1:20])
```

One Sample t-test

```
data: x[1:20]
t = 3.8245, df = 19, p-value = 0.001144
alternative hypothesis: true mean is not equal to 0
95 percent confidence interval:
0.3541055 1.2101894
sample estimates:
mean of x
0.7821474
```

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13.5.2. two-sample

```
x = rnorm(10,0.5)
y = rnorm(10,-0.5)
t.test(x,y)
```

```
Welch Two Sample t-test

data: x and y
t = 3.4296, df = 17.926, p-value = 0.003003
alternative hypothesis: true difference in means is not equal to 0
95 percent confidence interval:
 0.5811367 2.4204048
sample estimates:
 mean of x  mean of y
 0.7039205 -0.7968502
```

13.5.3. from a data.frame

In some situations, you may have data and groups as columns in a data.frame. See the following data.frame, for example

```
df = data.frame(value=c(x,y),group=as.factor(rep(c('g1','g2'),each=10)))
df
```

	value	group
1	1.12896674	g1
2	-1.26838101	g1
3	1.04577597	g1
4	1.69075585	g1
5	0.18672204	g1
6	1.99715092	g1
7	1.15424947	g1
8	0.37671442	g1
9	-0.09565723	g1
10	0.82290783	g1
11	-1.48530261	g2

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```
12 -1.29200440    g2
13 -0.18778362    g2
14  0.59205742    g2
15 -2.10065248    g2
16 -0.29961560    g2
17 -0.38985115    g2
18 -2.47126235    g2
19 -0.63654380    g2
20  0.30245611    g2
```

R allows us to perform a t-test using the `formula` notation.

```
t.test(value ~ group, data=df)
```

```
Welch Two Sample t-test

data: value by group
t = 3.4296, df = 17.926, p-value = 0.003003
alternative hypothesis: true difference in means between group g1 and group g2 is not equal to
95 percent confidence interval:
0.5811367 2.4204048
sample estimates:
mean in group g1 mean in group g2
0.7039205      -0.7968502
```

You read that as **value is a function of group**. In practice, this will do a t-test between the values in g1 vs g2.

13.5.4. Equivalence to linear model

```
t.test(value ~ group, data=df, var.equal=TRUE)
```

```
Two Sample t-test

data: value by group
```

13. The t-statistic and t-distribution

```
t = 3.4296, df = 18, p-value = 0.002989
alternative hypothesis: true difference in means between group g1 and group g2 is not equal to
95 percent confidence interval:
0.5814078 2.4201337
sample estimates:
mean in group g1 mean in group g2
0.7039205      -0.7968502
```

This is *equivalent* to:

```
res = lm(value ~ group, data=df)
summary(res)
```

```
Call:
lm(formula = value ~ group, data = df)

Residuals:
    Min      1Q  Median      3Q     Max 
-1.9723 -0.5600  0.2511  0.5252  1.3889 

Coefficients:
            Estimate Std. Error t value Pr(>|t|)    
(Intercept)  0.7039     0.3094   2.275  0.03538 *  
groupg2     -1.5008     0.4376  -3.430  0.00299 ** 
---
Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

Residual standard error: 0.9785 on 18 degrees of freedom
Multiple R-squared:  0.3952,    Adjusted R-squared:  0.3616 
F-statistic: 11.76 on 1 and 18 DF,  p-value: 0.002989
```

13.6. Power calculations

The power of a statistical test is the probability that the test will reject the null hypothesis when the alternative hypothesis is true. In other words, the power of a statistical test is the probability of not making a Type II error. The power of a

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statistical test depends on the significance level (alpha), the sample size, and the effect size.

The `power.t.test` function can be used to calculate the power of a one-sample t-test.

Looking at `help("power.t.test")`, we see that the function takes the following arguments:

- `n` - sample size
- `delta` - effect size
- `sd` - standard deviation of the sample
- `sig.level` - significance level
- `power` - power

We need to supply four of these arguments to calculate the fifth. For example, if we want to calculate the power of a one-sample t-test with a sample size of 5, a standard deviation of 1, and an effect size of 1, we can use the following command:

```
power.t.test(n = 5, delta = 1, sd = 1, sig.level = 0.05)
```

```
Two-sample t test power calculation
```

```
  n = 5
  delta = 1
  sd = 1
  sig.level = 0.05
  power = 0.2859276
  alternative = two.sided
```

NOTE: `n` is number in *each* group

This gives a nice summary of the power calculation. We can also extract the power value from the result:

```
power.t.test(n = 5, delta = 1, sd = 1,
             sig.level = 0.05, type='one.sample')$power
```

```
[1] 0.4013203
```

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💡 Tip

When getting results from a function that don't look "computable" such as those from `power.t.test`, you can use the `$` operator to extract the value you want. In this case, we want the `power` value from the result of `power.t.test`. How would you know what to extract? You can use the `names` function or the `str` function to see the structure of the result. For example:

```
names(power.t.test(n = 5, delta = 1, sd = 1,
                    sig.level = 0.05, type='one.sample'))  
  
[1] "n"           "delta"        "sd"           "sig.level"    "power"  
[6] "alternative" "note"         "method"  
  
# or  
str(power.t.test(n = 5, delta = 1, sd = 1,
                  sig.level = 0.05, type='one.sample'))  
  
List of 8  
 $ n          : num 5  
 $ delta      : num 1  
 $ sd         : num 1  
 $ sig.level   : num 0.05  
 $ power       : num 0.401  
 $ alternative: chr "two.sided"  
 $ note        : NULL  
 $ method      : chr "One-sample t test power calculation"  
 - attr(*, "class")= chr "power.htest"
```

Alternatively, we may know a lot about our experimental system and want to calculate the sample size needed to achieve a certain power. For example, if we want to achieve a power of 0.8 with a standard deviation of 1 and an effect size of 1, we can use the following command:

```
power.t.test(delta = 1, sd = 1, sig.level = 0.05, power = 0.8, type = "one.sample")
```

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```
One-sample t test power calculation
```

```
n = 9.937864
delta = 1
sd = 1
sig.level = 0.05
power = 0.8
alternative = two.sided
```

The `power.t.test` function is convenient and quite fast. As we've seen before, though, sometimes the distribution of the test statistics is now easily calculated. In those cases, we can use simulation to calculate the power of a statistical test. For example, if we want to calculate the power of a one-sample t-test with a sample size of 5, a standard deviation of 1, and an effect size of 1, we can use the following command:

```
sim_t_test_pval <- function(n = 5, delta = 1, sd = 1, sig.level = 0.05) {
  x = rnorm(n, delta, sd)
  t.test(x)$p.value <= sig.level
}
pow = mean(replicate(1000, sim_t_test_pval()))
pow
```

```
[1] 0.405
```

Let's break this down. First, we define a function called `sim_t_test_pval` that takes the same arguments as the `power.t.test` function. Inside the function, we simulate a sample of size `n` from a normal distribution with mean `delta` and standard deviation `sd`. Then, we perform a one-sample t-test on the sample and return a logical value indicating whether the p-value is less than the significance level. Next, we use the `replicate` function to repeat the simulation 1000 times. Finally, we calculate the proportion of simulations in which the p-value was less than the significance level. This proportion is an estimate of the power of the one-sample t-test.

Let's compare the results of the `power.t.test` function and our simulation-based approach:

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```
power.t.test(n = 5, delta = 1, sd = 1, sig.level = 0.05, type='one.sample')$power  
[1] 0.4013203  
  
mean(replicate(1000, sim_t_test_pval(n = 5, delta = 1, sd = 1, sig.level = 0.05)))  
[1] 0.414
```

13.7. Resources

See the [pwr package](#) for more information on power calculations.

14. K-means clustering

14.1. History of the k-means algorithm

The k-means clustering algorithm was first proposed by Stuart Lloyd in 1957 as a technique for pulse-code modulation. However, it was not published until 1982. In 1965, Edward W. Forgy published an essentially identical method, which became widely known as the k-means algorithm. Since then, k-means clustering has become one of the most popular unsupervised learning techniques in data analysis and machine learning.

K-means clustering is a method for finding patterns or groups in a dataset. It is an unsupervised learning technique, meaning that it doesn't rely on previously labeled data for training. Instead, it identifies structures or patterns directly from the data based on the similarity between data points (see Figure 14.1).

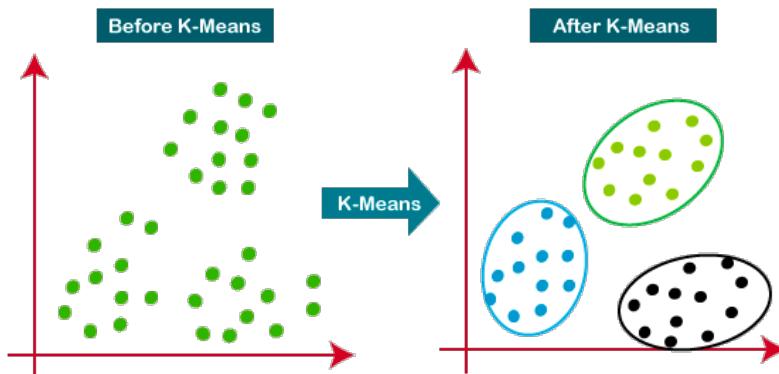


Figure 14.1.: K-means clustering takes a dataset and divides it into k clusters.

In simple terms, k-means clustering aims to divide a dataset into k distinct groups or clusters, where each data point belongs to the cluster with the nearest mean (average). The goal is to minimize the variability within each cluster while maximizing the differences between clusters. This helps to reveal hidden

14. K-means clustering

patterns or relationships in the data that might not be apparent otherwise.

14.2. The k-means algorithm

The k-means algorithm follows these general steps:

1. Choose the number of clusters k .
2. Initialize the cluster centroids randomly by selecting k data points from the dataset.
3. Assign each data point to the nearest centroid.
4. Update the centroids by computing the mean of all the data points assigned to each centroid.
5. Repeat steps 3 and 4 until the centroids no longer change or a certain stopping criterion is met (e.g., a maximum number of iterations).

The algorithm converges when the centroids stabilize or no longer change significantly. The final clusters represent the underlying patterns or structures in the data. Advantages and disadvantages of k-means clustering

14.3. Pros and cons of k-means clustering

Compared to other clustering algorithms, k-means has several advantages:

- **Simplicity and ease of implementation** The k-means algorithm is relatively straightforward and can be easily implemented, even for large datasets.
- **Scalability** The algorithm can be adapted for large datasets using various optimization techniques or parallel processing.
- **Speed** K-means is generally faster than other clustering algorithms, especially when the number of clusters k is small.

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- **Interpretability** The results of k-means clustering are easy to understand, as the algorithm assigns each data point to a specific cluster based on its similarity to the cluster's centroid.

However, k-means clustering has several disadvantages as well:

- **Choice of k** Selecting the appropriate number of clusters can be challenging and often requires domain knowledge or experimentation. A poor choice of k may yield poor results.
- **Sensitivity to initial conditions** The algorithm's results can vary depending on the initial placement of centroids. To overcome this issue, the algorithm can be run multiple times with different initializations and the best solution can be chosen based on a criterion (e.g., minimizing within-cluster variation).
- **Assumes spherical clusters** K-means assumes that clusters are spherical and evenly sized, which may not always be the case in real-world datasets. This can lead to poor performance if the underlying clusters have different shapes or densities.
- **Sensitivity to outliers** The algorithm is sensitive to outliers, which can heavily influence the position of centroids and the final clustering result. Preprocessing the data to remove or mitigate the impact of outliers can help improve the performance of k-means clustering.

Despite limitations, k-means clustering remains a popular and widely used method for exploring and analyzing data, particularly in biological data analysis, where identifying patterns and relationships can provide valuable insights into complex systems and processes.

14.4. An example of k-means clustering

14.4.1. The data and experimental background

The data we are going to use are from DeRisi, Iyer, and Brown (1997). From their abstract:

DNA microarrays containing virtually every gene of *Saccharomyces cerevisiae* were used to carry out a comprehensive investigation of the temporal program of gene expression accompanying the metabolic shift from fermentation to respiration. The expression profiles observed for genes with known metabolic functions pointed to features of the metabolic reprogramming that occur during the diauxic shift, and the expression patterns of many previously uncharacterized genes provided clues to their possible functions.

These data are available from NCBI GEO as [GSE28](#).

In the case of the baker's or brewer's yeast *Saccharomyces cerevisiae* growing on glucose with plenty of aeration, the diauxic growth pattern is commonly observed in batch culture. During the first growth phase, when there is plenty of glucose and oxygen available, the yeast cells prefer glucose fermentation to aerobic respiration even though aerobic respiration is the more efficient pathway to grow on glucose. This experiment profiles gene expression for 6400 genes over a time course during which the cells are undergoing a [diauxic shift](#).

The data in deRisi et al. have no replicates and are time course data. Sometimes, seeing how groups of genes behave can give biological insight into the experimental system or the function of individual genes. We can use clustering to group genes that have a similar expression pattern over time and then potentially look at the genes that do so.

Our goal, then, is to use `kmeans` clustering to divide highly variable (informative) genes into groups and then to visualize those groups.

14.5. Getting data

These data were deposited at NCBI GEO back in 2002. GEO-query can pull them out easily.

```
library(GEOquery)
gse = getGEO("GSE28")[[1]]
class(gse)
```

```
[1] "ExpressionSet"
attr(,"package")
[1] "Biobase"
```

GEOquery is a little dated and was written before the SummarizedExperiment existed. However, Bioconductor makes a conversion from the old ExpressionSet that GEOquery uses to the SummarizedExperiment that we see so commonly used now.

```
library(SummarizedExperiment)
gse = as(gse, "SummarizedExperiment")
gse
```

```
class: SummarizedExperiment
dim: 6400 7
metadata(3): experimentData annotation protocolData
assays(1): exprs
rownames(6400): 1 2 ... 6399 6400
rowData names(20): ID ORF ... FAILED IS_CONTAMINATED
colnames(7): GSM887 GSM888 ... GSM892 GSM893
colData names(33): title geo_accession ... supplementary_file
  data_row_count
```

Taking a quick look at the `colData()`, it might be that we want to reorder the columns a bit.

```
colData(gse)$title
```

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```
[1] "diauxic shift timecourse: 15.5 hr" "diauxic shift timecourse: 0 hr"  
[3] "diauxic shift timecourse: 18.5 hr" "diauxic shift timecourse: 9.5 hr"  
[5] "diauxic shift timecourse: 11.5 hr" "diauxic shift timecourse: 13.5 hr"  
[7] "diauxic shift timecourse: 20.5 hr"
```

So, we can reorder by hand to get the time course correct:

```
gse = gse[, c(2,4,5,6,1,3,7)]
```

14.6. Preprocessing

In gene expression data analysis, the primary objective is often to identify genes that exhibit significant differences in expression levels across various conditions, such as diseased vs. healthy samples or different time points in a time-course experiment. However, gene expression datasets are typically large, noisy, and contain numerous genes that do not exhibit substantial changes in expression levels. Analyzing all genes in the dataset can be computationally intensive and may introduce noise or false positives in the results.

One common approach to reduce the complexity of the dataset and focus on the most informative genes is to subset the genes based on their standard deviation in expression levels across the samples. The standard deviation is a measure of dispersion or variability in the data, and genes with high standard deviations have more variation in their expression levels across the samples.

By selecting genes with high standard deviations, we focus on genes that show relatively large changes in expression levels across different conditions. These genes are more likely to be biologically relevant and involved in the underlying processes or pathways of interest. In contrast, genes with low standard deviations exhibit little or no change in expression levels and are less likely to be informative for the analysis. It turns out that applying filtering based on criteria such as standard deviation can also increase power and reduce false positives in the analysis (Bourgon, Gentleman, and Huber 2010).

14. K-means clustering

To subset the genes for analysis based on their standard deviation, the following steps can be followed: Calculate the standard deviation of each gene's expression levels across all samples. Set a threshold for the standard deviation, which can be determined based on domain knowledge, data distribution, or a specific percentile of the standard deviation values (e.g., selecting the top 10% or 25% of genes with the highest standard deviations). Retain only the genes with a standard deviation above the chosen threshold for further analysis.

By subsetting the genes based on their standard deviation, we can reduce the complexity of the dataset, speed up the subsequent analysis, and increase the likelihood of detecting biologically meaningful patterns and relationships in the gene expression data. The threshold for the standard deviation cutoff is rather arbitrary, so it may be beneficial to try a few to check for sensitivity of findings.

```
sds = apply(assays(gse)[[1]], 1, sd)
hist(sds)
```

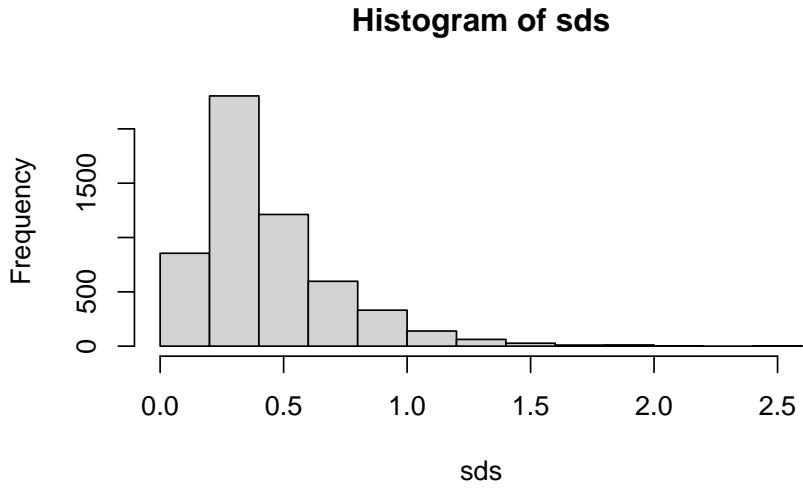


Figure 14.2.: Histogram of standard deviations for all genes in the deRisi dataset.

Examining the plot, we can see that the most highly variable genes have an $sd > 0.8$ or so (arbitrary). We can, for convenience, create a new `SummarizedExperiment` that contains only our most highly variable genes.

14. K-means clustering

```
idx = sds>0.8 & !is.na(sds)
gse_sub = gse[idx,]
```

14.7. Clustering

Now, `gse_sub` contains a subset of our data.

The `kmeans` function takes a matrix and the number of clusters as arguments.

```
k = 4
km = kmeans(assays(gse_sub)[[1]], 4)
```

The `km` kmeans result contains a vector, `km$cluster`, which gives the cluster associated with each gene. We can plot the genes for each cluster to see how these different genes behave.

```
expression_values = assays(gse_sub)[[1]]
par(mfrow=c(2,2), mar=c(3,4,1,2)) # this allows multiple plots per page
for(i in 1:k) {
  matplot(t(expression_values[km$cluster==i, ]), type='l', ylim=c(-3,3),
          ylab = paste("cluster", i))
}
```

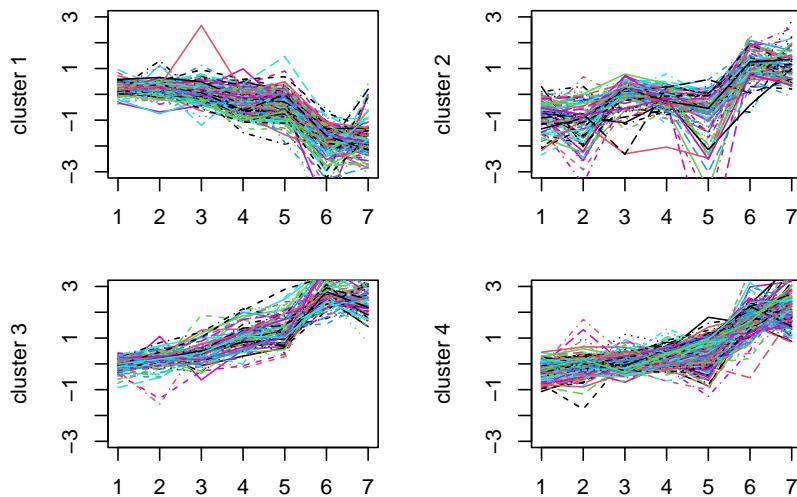


Figure 14.3.: Gene expression profiles for the four clusters identified by k-means clustering. Each line represents a gene in the cluster, and each column represents a time point in the experiment. Each cluster shows a distinct trend where the genes in the cluster are potentially co-regulated.

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Try this with different size k. Perhaps go back to choose more genes (using a smaller cutoff for sd).

14.8. Summary

In this lesson, we have learned how to use k-means clustering to identify groups of genes that behave similarly over time. We have also learned how to subset our data to focus on the most informative genes.

Part V.

Bioconductor

15. Accessing and working with public omics data

15.1. Background

The data we are going to access are from [this paper](#).

Background: The tumor microenvironment is an important factor in cancer immunotherapy response. To further understand how a tumor affects the local immune system, we analyzed immune gene expression differences between matching normal and tumor tissue.

Methods: We analyzed public and new gene expression data from solid cancers and isolated immune cell populations. We also determined the correlation between CD8, FoxP3 IHC, and our gene signatures.

Results: We observed that regulatory T cells (Tregs) were one of the main drivers of immune gene expression differences between normal and tumor tissue. A tumor-specific CD8 signature was slightly lower in tumor tissue compared with normal of most (12 of 16) cancers, whereas a Treg signature was higher in tumor tissue of all cancers except liver. Clustering by Treg signature found two groups in colorectal cancer datasets. The high Treg cluster had more samples that were consensus molecular subtype 1/4, right-sided, and microsatellite-instable, compared with the low Treg cluster. Finally, we found that the correlation between signature and IHC was low in our small dataset, but samples in the high Treg cluster had significantly more CD8+ and FoxP3+ cells compared with the low Treg cluster.

Conclusions: Treg gene expression is highly

15. Accessing and working with public omics data

indicative of the overall tumor immune environment. Impact: In comparison with the consensus molecular subtype and microsatellite status, the Treg signature identifies more colorectal tumors with high immune activation that may benefit from cancer immunotherapy.

In this little exercise, we will:

1. Access public omics data using the `GEOquery` package
2. Get an opportunity to work with another `SummarizedExperiment` object.
3. Perform a simple unsupervised analysis to visualize these public data.

15.2. GEOquery to multidimensional scaling

The first step is to install the `R` package `GEOquery`. This package allows us to access data from the Gene Expression Omnibus (GEO) database. GEO is a public repository of omics data.

```
BiocManager::install("GEOquery")
```

`GEOquery` has only one commonly used function, `getGEO()` which takes a GEO accession number as an argument. The GEO accession number is a unique identifier for a dataset.

Use the `GEOquery` package to fetch data about [GSE103512](#).

```
library(GEOquery)
gse = getGEO("GSE103512")[[1]]
```

The first step, a detail, is to convert from the older Bioconductor data structure (`GEOquery` was written in 2007), the `ExpressionSet`, to the newer `SummarizedExperiment`.

```
library(SummarizedExperiment)
se = as(gse, "SummarizedExperiment")
```

15. Accessing and working with public omics data

- What is the class of `se`?
- What are the dimensions of `se`?
- What are the dimensions of the `assay` slot of `se`?
- What are the dimensions of the `colData` slot of `se`?
- What variables are in the `colData` slot of `se`?

Examine two variables of interest, cancer type and tumor/normal status. The `with` function is a convenience to allow us to access variables in a data frame by name (rather than having to do `dataframe$variable_name`.

```
with(colData(se),table(`cancer.type.ch1`,`normal.ch1`))
```

		normal.ch1
cancer.type.ch1	no	yes
BC	65	10
CRC	57	12
NSCLC	60	9
PCA	60	7

Filter gene expression by variance to find most informative genes. It is common practice to filter genes by standard deviation or some other measure of variability and keep the top X percent of them when performing dimensionality reduction. There is not a single right answer to what percentage to use, so try a few to see what happens.

In the example code, I chose to use the top 500 genes by standard deviation.

```
sds = apply(assay(se, 'exprs'), 1, sd)
dat = assay(se, 'exprs')[order(sds, decreasing = TRUE)[1:500],]
```

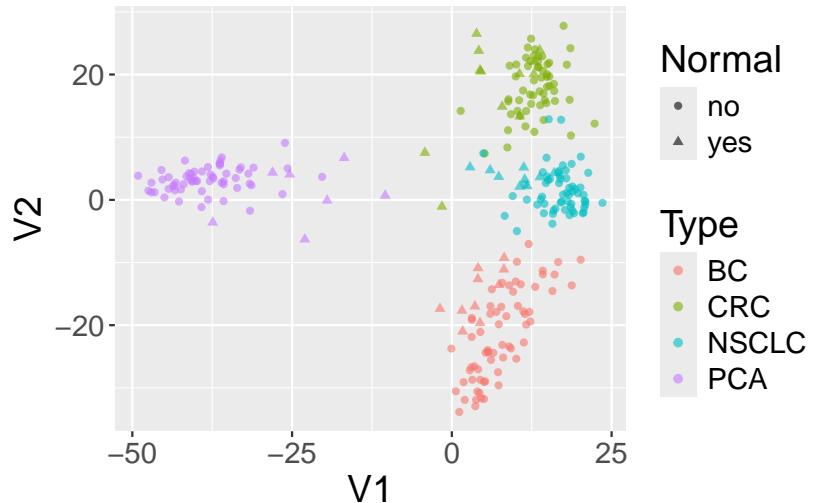
Perform [multidimensional scaling](#) and prepare for plotting. We will be using `ggplot2`, so we need to make a `data.frame` before plotting.

```
mdsvals = cmdscale(dist(t(dat)))
mdsvals = as.data.frame(mdsvals)
mdsvals>Type=factor(colData(se)[,'cancer.type.ch1'])
mdsvals$Normal = factor(colData(se)[,'normal.ch1'])
```

15. Accessing and working with public omics data

And do the plot.

```
library(ggplot2)
ggplot(mdsvals, aes(x=V1,y=V2,shape=Normal,color=Type)) +
  geom_point( alpha=0.6) + theme(text=element_text(size = 18))
```



- What do you see?

16. Introduction to SummarizedExperiment

The `SummarizedExperiment` class is used to store rectangular matrices of experimental results, which are commonly produced by sequencing and microarray experiments. Each object stores observations of one or more samples, along with additional meta-data describing both the observations (features) and samples (phenotypes).

A key aspect of the `SummarizedExperiment` class is the coordination of the meta-data and assays when subsetting. For example, if you want to exclude a given sample you can do for both the meta-data and assay in one operation, which ensures the meta-data and observed data will remain in sync. Improperly accounting for meta and observational data has resulted in a number of incorrect results and retractions so this is a very desirable property.

`SummarizedExperiment` is in many ways similar to the historical `ExpressionSet`, the main distinction being that `SummarizedExperiment` is more flexible in its row information, allowing both `GRanges` based as well as those described by arbitrary `DataFrames`. This makes it ideally suited to a variety of experiments, particularly sequencing based experiments such as RNA-Seq and ChIP-Seq.

```
BiocManager::install('airway')
BiocManager::install('SummarizedExperiment')
```

16.1. Anatomy of a `SummarizedExperiment`

The *SummarizedExperiment* package contains two classes: `SummarizedExperiment` and `RangedSummarizedExperiment`.

16. Introduction to *SummarizedExperiment*

SummarizedExperiment is a matrix-like container where rows represent features of interest (e.g. genes, transcripts, exons, etc.) and columns represent samples. The objects contain one or more assays, each represented by a matrix-like object of numeric or other mode. The rows of a **SummarizedExperiment** object represent features of interest. Information about these features is stored in a **DataFrame** object, accessible using the function **rowData()**. Each row of the **DataFrame** provides information on the feature in the corresponding row of the **SummarizedExperiment** object. Columns of the **DataFrame** represent different attributes of the features of interest, e.g., gene or transcript IDs, etc.

RangedSummarizedExperiment is the “child” of the **SummarizedExperiment** class which means that all the methods on **SummarizedExperiment** also work on a **RangedSummarizedExperiment**.

The fundamental difference between the two classes is that the rows of a **RangedSummarizedExperiment** object represent genomic ranges of interest instead of a **DataFrame** of features. The **RangedSummarizedExperiment** ranges are described by a **GRanges** or a **GRangesList** object, accessible using the **rowRanges()** function.

Figure 16.1 displays the class geometry and highlights the vertical (column) and horizontal (row) relationships.

16.1.1. Assays

The **airway** package contains an example dataset from an RNA-Seq experiment of read counts per gene for airway smooth muscles. These data are stored in a **RangedSummarizedExperiment** object which contains 8 different experimental and assays 64,102 gene transcripts.

```
Loading required package: airway
```

16. Introduction to SummarizedExperiment

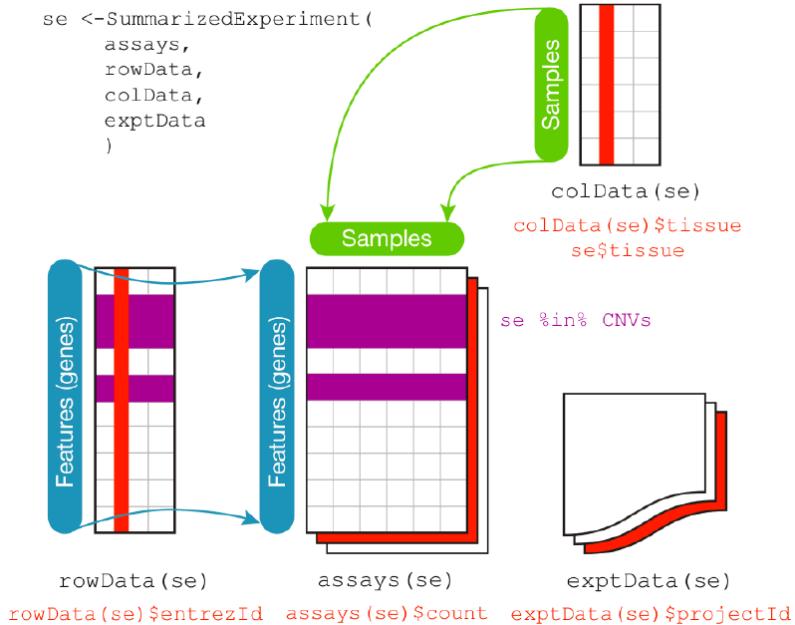


Figure 16.1.: Summarized Experiment. There are three main components, the `colData()`, the `rowData()` and the `assays()`. The accessors for the various parts of a complete `SummarizedExperiment` object match the names.

```

library(SummarizedExperiment)
data(airway, package="airway")
se <- airway
se

```

```

class: RangedSummarizedExperiment
dim: 63677 8
metadata(1): ''
assays(1): counts
rownames(63677): ENSG00000000003 ENSG00000000005 ... ENSG00000273492
  ENSG00000273493
rowData names(10): gene_id gene_name ... seq_coord_system symbol
colnames(8): SRR1039508 SRR1039509 ... SRR1039520 SRR1039521
colData names(9): SampleName cell ... Sample BioSample

```

To retrieve the experiment data from a `SummarizedExperiment` object one can use the `assays()` accessor. An object can have multiple assay datasets each of which can be accessed using the `$` operator. The `airway` dataset contains only one assay (`counts`). Here each row represents a gene transcript and each column one of the samples.

```
assays(se)$counts
```

	SRR103580	SRR908520	SRR2035830	SRR6035870	SRR703520	039521
ENSG000007900000318	873	408	1138	1047	770	572
ENSG0000000000050	0	0	0	0	0	0
ENSG000007004595	621	365	587	799	417	508
ENSG00002000045711	263	164	245	331	233	229
ENSG000000004655	40	35	78	63	76	60
ENSG000000009380	2	0	1	0	0	0
ENSG0000251003679	6177	4252	6721	11027	5176	7995
ENSG0000000010362	1733	881	1424	1439	1359	1109
ENSG0000000010380	595	493	820	714	696	704
ENSG0000000011236	464	175	658	584	360	269

16.1.2. ‘Row’ (regions-of-interest) data

The `rowRanges()` accessor is used to view the range information for a `RangedSummarizedExperiment`. (Note if this were the parent `SummarizedExperiment` class we’d use `rowData()`). The data are stored in a `GRangesList` object, where each list element corresponds to one gene transcript and the ranges in each `GRanges` correspond to the exons in the transcript.

```
rowRanges(se)
```

```
GRangesList object of length 63677:
$ENSG000000000003
GRanges object with 17 ranges and 2 metadata columns:
  seqnames      ranges strand |  exon_id    exon_name
  <Rle>      <IRanges> <Rle> | <integer>  <character>
 [1]      X 99883667-99884983     - |    667145 ENSE00001459322
 [2]      X 99885756-99885863     - |    667146 ENSE00000868868
 [3]      X 99887482-99887565     - |    667147 ENSE00000401072
 [4]      X 99887538-99887565     - |    667148 ENSE00001849132
 [5]      X 99888402-99888536     - |    667149 ENSE00003554016
 ...
[13]      ...          ...     ... |    ...
[13]      X 99890555-99890743     - |    667156 ENSE00003512331
```

16. Introduction to *SummarizedExperiment*

```
[14]      X 99891188-99891686      - | 667158 ENSE00001886883
[15]      X 99891605-99891803      - | 667159 ENSE00001855382
[16]      X 99891790-99892101      - | 667160 ENSE00001863395
[17]      X 99894942-99894988      - | 667161 ENSE00001828996
-----
seqinfo: 722 sequences (1 circular) from an unspecified genome

...
<63676 more elements>
```

16.1.3. ‘Column’ (sample) data

Sample meta-data describing the samples can be accessed using `colData()`, and is a `DataFrame` that can store any number of descriptive columns for each sample row.

```
colData(se)
```

```
DataFrame with 8 rows and 9 columns
  SampleName      cell      dex    albut      Run avgLength
  <factor> <factor> <factor> <factor> <factor> <integer>
SRR1039508 GSM1275862 N61311    untrt    untrt SRR1039508     126
SRR1039509 GSM1275863 N61311    trt      untrt SRR1039509     126
SRR1039512 GSM1275866 N052611   untrt    untrt SRR1039512     126
SRR1039513 GSM1275867 N052611   trt      untrt SRR1039513      87
SRR1039516 GSM1275870 N080611   untrt    untrt SRR1039516     120
SRR1039517 GSM1275871 N080611   trt      untrt SRR1039517     126
SRR1039520 GSM1275874 N061011   untrt    untrt SRR1039520     101
SRR1039521 GSM1275875 N061011   trt      untrt SRR1039521      98
  Experiment      Sample      BioSample
  <factor> <factor> <factor>
SRR1039508 SRX384345 SRS508568 SAMN02422669
SRR1039509 SRX384346 SRS508567 SAMN02422675
SRR1039512 SRX384349 SRS508571 SAMN02422678
SRR1039513 SRX384350 SRS508572 SAMN02422670
SRR1039516 SRX384353 SRS508575 SAMN02422682
SRR1039517 SRX384354 SRS508576 SAMN02422673
SRR1039520 SRX384357 SRS508579 SAMN02422683
SRR1039521 SRX384358 SRS508580 SAMN02422677
```

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This sample metadata can be accessed using the `$` accessor which makes it easy to subset the entire object by a given phenotype.

```
# subset for only those samples treated with dexamethasone  
se[, se$dex == "trt"]
```

```
class: RangedSummarizedExperiment  
dim: 63677 4  
metadata(1): ''  
assays(1): counts  
rownames(63677): ENSG00000000003 ENSG00000000005 ... ENSG00000273492  
ENSG00000273493  
rowData names(10): gene_id gene_name ... seq_coord_system symbol  
colnames(4): SRR1039509 SRR1039513 SRR1039517 SRR1039521  
colData names(9): SampleName cell ... Sample BioSample
```

16.1.4. Experiment-wide metadata

Meta-data describing the experimental methods and publication references can be accessed using `metadata()`.

```
metadata(se)
```

```
[[1]]
```

```
Experiment data
```

```
  Experimenter name: Himes BE
```

```
  Laboratory: NA
```

```
  Contact information:
```

```
  Title: RNA-Seq transcriptome profiling identifies CRISPLD2 as a glucocorticoid responsive gene
```

```
  URL: http://www.ncbi.nlm.nih.gov/pubmed/24926665
```

```
  PMIDs: 24926665
```

```
Abstract: A 226 word abstract is available. Use 'abstract' method.
```

Note that `metadata()` is just a simple list, so it is appropriate for *any* experiment wide metadata the user wishes to save, such as storing model formulas.

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```
metadata(se)$formula <- counts ~ dex + albut  
  
metadata(se)  
  
[[1]]  
Experiment data  
  Experimenter name: Himes BE  
  Laboratory: NA  
  Contact information:  
    Title: RNA-Seq transcriptome profiling identifies CRISPLD2 as a glucocorticoid responsive gene  
    URL: http://www.ncbi.nlm.nih.gov/pubmed/24926665  
    PMIDs: 24926665  
  
  Abstract: A 226 word abstract is available. Use 'abstract' method.  
  
$formula  
counts ~ dex + albut
```

16.2. Common operations on *SummarizedExperiment*

16.2.1. Subsetting

- [Performs two dimensional subsetting, just like subsetting a matrix or data frame.

```
# subset the first five transcripts and first three samples  
se[1:5, 1:3]
```

```
class: RangedSummarizedExperiment  
dim: 5 3  
metadata(2): '' formula  
assays(1): counts  
rownames(5): ENSG000000000003 ENSG000000000005 ENSG000000000419  
  ENSG00000000457 ENSG00000000460  
rowData names(10): gene_id gene_name ... seq_coord_system symbol  
colnames(3): SRR1039508 SRR1039509 SRR1039512  
colData names(9): SampleName cell ... Sample BioSample
```

16. Introduction to *SummarizedExperiment*

- `$` operates on `colData()` columns, for easy sample extraction.

```
se[, se$cell == "N61311"]
```

```
class: RangedSummarizedExperiment
dim: 63677 2
metadata(2): '' formula
assays(1): counts
rownames(63677): ENSG00000000003 ENSG00000000005 ... ENSG00000273492
  ENSG00000273493
rowData names(10): gene_id gene_name ... seq_coord_system symbol
colnames(2): SRR1039508 SRR1039509
colData names(9): SampleName cell ... Sample BioSample
```

16.2.2. Getters and setters

- `rowRanges()` / (`rowData()`, `colData()`, `metadata()`)

```
counts <- matrix(1:15, 5, 3, dimnames=list(LETTERS[1:5], LETTERS[1:3]))

dates <- SummarizedExperiment(assays=list(counts=counts),
                             rowData=DataFrame(month=month.name[1:5], day=1:5))

# Subset all January assays
dates[rowData(dates)$month == "January", ]
```

```
class: SummarizedExperiment
dim: 1 3
metadata(0):
assays(1): counts
rownames(1): A
rowData names(2): month day
colnames(3): A B C
colData names(0):
```

16. Introduction to *SummarizedExperiment*

- `assay()` versus `assays()` There are two accessor functions for extracting the assay data from a `SummarizedExperiment` object. `assays()` operates on the entire list of assay data as a whole, while `assay()` operates on only one assay at a time. `assay(x, i)` is simply a convenience function which is equivalent to `assays(x)[[i]]`.

```
assays(se)
```

```
List of length 1  
names(1): counts
```

```
assays(se)[[1]][1:5, 1:5]
```

	SRR1039508	SRR1039509	SRR1039512	SRR1039513	SRR1039516
ENSG000000000003	679	448	873	408	1138
ENSG000000000005	0	0	0	0	0
ENSG00000000419	467	515	621	365	587
ENSG00000000457	260	211	263	164	245
ENSG00000000460	60	55	40	35	78

```
# assay defaults to the first assay if no i is given  
assay(se)[1:5, 1:5]
```

	SRR1039508	SRR1039509	SRR1039512	SRR1039513	SRR1039516
ENSG000000000003	679	448	873	408	1138
ENSG000000000005	0	0	0	0	0
ENSG00000000419	467	515	621	365	587
ENSG00000000457	260	211	263	164	245
ENSG00000000460	60	55	40	35	78

```
assay(se, 1)[1:5, 1:5]
```

	SRR1039508	SRR1039509	SRR1039512	SRR1039513	SRR1039516
ENSG000000000003	679	448	873	408	1138
ENSG000000000005	0	0	0	0	0
ENSG00000000419	467	515	621	365	587
ENSG00000000457	260	211	263	164	245
ENSG00000000460	60	55	40	35	78

16.2.3. Range-based operations

- `subsetByOverlaps()` *SummarizedExperiment* objects support all of the `findOverlaps()` methods and associated functions. This includes `subsetByOverlaps()`, which makes it easy to subset a *SummarizedExperiment* object by an interval.

In the next code block, we define a region of interest (or many regions of interest) and then subset our *SummarizedExperiment* by overlaps with this region.

```
# Subset for only rows which are in the interval 100,000 to 110,000 of
# chromosome 1
roi <- GRanges(seqnames="1", ranges=100000:1100000)
sub_se = subsetByOverlaps(se, roi)
sub_se

class: RangedSummarizedExperiment
dim: 74 8
metadata(2): '' formula
assays(1): counts
rownames(74): ENSG00000131591 ENSG00000177757 ... ENSG00000272512
  ENSG00000273443
rowData names(10): gene_id gene_name ... seq_coord_system symbol
colnames(8): SRR1039508 SRR1039509 ... SRR1039520 SRR1039521
colData names(9): SampleName cell ... Sample BioSample

dim(sub_se)

[1] 74 8
```

16.3. Constructing a *SummarizedExperiment*

Often, *SummarizedExperiment* or *RangedSummarizedExperiment* objects are returned by functions written by other packages. However it is possible to create them by hand with a call to the `SummarizedExperiment()` constructor. The code below is

16. Introduction to *SummarizedExperiment*

simply to illustrate the mechanics of creating an object from scratch. In practice, you will probably have the pieces of the object from other sources such as Excel files or csv files.

Constructing a `RangedSummarizedExperiment` with a `GRanges` as the `rowRanges` argument:

```
nrows <- 200
ncols <- 6
counts <- matrix(runif(nrows * ncols, 1, 1e4), nrows)
rowRanges <- GRanges(rep(c("chr1", "chr2"), c(50, 150)),
                      IRanges(floor(runif(200, 1e5, 1e6)), width=100),
                      strand=sample(c("+", "-"), 200, TRUE),
                      feature_id=sprintf("ID%03d", 1:200))
colData <- DataFrame(Treatment=rep(c("ChIP", "Input"), 3),
                      row.names=LETTERS[1:6])

SummarizedExperiment(assays=list(counts=counts),
                     rowRanges=rowRanges, colData=colData)
```

```
class: RangedSummarizedExperiment
dim: 200 6
metadata(0):
assays(1): counts
rownames: NULL
rowData names(1): feature_id
colnames(6): A B ... E F
colData names(1): Treatment
```

A `SummarizedExperiment` can be constructed with or without supplying a `DataFrame` for the `rowData` argument:

```
SummarizedExperiment(assays=list(counts=counts), colData=colData)
```

```
class: SummarizedExperiment
dim: 200 6
metadata(0):
assays(1): counts
rownames: NULL
rowData names(0):
```

16. Introduction to SummarizedExperiment

```
colnames(6): A B ... E F  
colData names(1): Treatment
```

17. EDA with PCA

17.1. Introduction

In this tutorial, we will use the GEOquery package to download a dataset from the Gene Expression Omnibus (GEO) and perform some exploratory data analysis (EDA) using principal components analysis (PCA).

17.2. Downloading data from GEO

The GEOquery package can be used to download data from GEO. The `getGEO` function takes a GEO accession number as an argument and returns a list of ExpressionSet objects. The `[[1]]` at the end of the `getGEO` call is used to extract the first (and only) ExpressionSet object from the list.

Historically, it was not uncommon for GEO datasets to contain multiple separate experiments. In those cases, the `[[1]]` would need to be replaced with the index of the experiment of interest. However, it is now uncommon for GEO datasets to contain multiple experiments, but the `[[1]]` is still needed to extract the ExpressionSet object from the list.

```
library(GEOquery)
library(SummarizedExperiment)
```

ExpressionSet objects are a type of Bioconductor object that is used to store gene expression data. The `as` function can be used to convert the ExpressionSet object to a SummarizedExperiment object, which is a newer Bioconductor object that is used to store gene expression data. The SummarizedExperiment object is preferred over the ExpressionSet object so we

17. EDA with PCA

immediately convert the ExpressionSet object to a SummarizedExperiment.

```
gse <- getGEO("GSE30219")[[1]]
```

Found 1 file(s)

GSE30219_series_matrix.txt.gz

```
se <- as(gse, "SummarizedExperiment")
```

17.3. Filtering genes

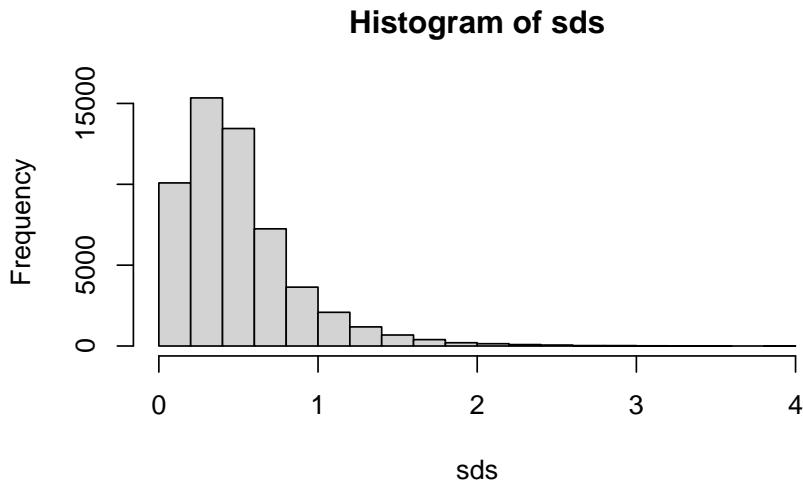
When performing PCA, it is common to filter to the most variable genes before performing the PCA. Limiting genes to the most variable genes can help to reduce the computational burden of the PCA.

We can calculate the standard deviation of each gene using the `apply` function. The `apply` function takes a matrix as the first argument and a 1 or 2 to indicate whether the function should be applied to the rows or columns of the matrix. The `sd` function calculates the standard deviation of a vector and is performed on each row of the matrix.

A histogram of the standard deviations is not that useful, but it is easy to make.

```
sds = apply(assay(se, 'exprs'), 1, sd)
hist(sds)
```

17. EDA with PCA



Here, we produce a subset of the `SummarizedExperiment` object that contains only the 500 most variable genes. We'll use this subset for the rest of the tutorial. Feel free to revisit the number of genes you choose to keep and see how it affects the PCA.

```
sub_se = se[order(sds,decreasing = TRUE)[1:500],]
```

17.4. PCA

PCA is a method for dimensionality reduction. It is a linear transformation that finds the directions of maximum variance in a dataset and projects it onto a new subspace with equal or fewer dimensions than the original one. The orthogonal axes (principal components) of the new subspace can be interpreted as the directions of maximum variance given the constraint that the new feature axes are orthogonal to each other.

17. EDA with PCA

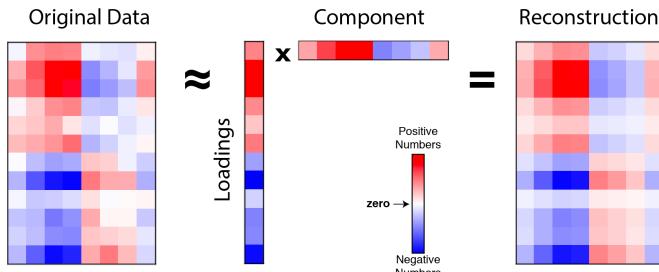


Figure 17.1.: The matrix decomposition of the first PC and how we can use it to construct the dimensionally-reduced dataset.

```
# read the help for prcomp here to see what the arguments are
# ?prcomp
pca = prcomp(t(assay(sub_se, 'exprs')))
```

The PCA algorithm results in a rotation matrix that can be used to transform the original data into the new subspace. The rotation matrix is stored in the `rotation` slot of the `prcomp` object and represents the *loadings* of each gene for each principle component. The `prcomp` function also stores the coordinates of the samples in the new subspace in the `x` slot, which represents the locations of the samples in principle component space.

```
str(pca)
```

```
List of 5
 $ sdev     : num [1:307] 27.01 22.78 13.46 10.43 9.35 ...
 $ rotation: num [1:500, 1:307] -0.1091 0.0598 -0.0474 -0.0513 -0.0903 ...
 ..- attr(*, "dimnames")=List of 2
 ...$ : chr [1:500] "209125_at" "209988_s_at" "223678_s_at" "218835_at" ...
 ...$ : chr [1:307] "PC1" "PC2" "PC3" "PC4" ...
 $ center   : Named num [1:500] 6.86 5.56 7.99 10.39 7.82 ...
 ..- attr(*, "names")= chr [1:500] "209125_at" "209988_s_at" "223678_s_at" "218835_at" ...
 $ scale    : logi FALSE
 $ x        : num [1:307, 1:307] 0.571 -3.528 5.289 -31.486 1.273 ...
 ..- attr(*, "dimnames")=List of 2
 ...$ : chr [1:307] "GSM748053" "GSM748054" "GSM748055" "GSM748056" ...
 ...$ : chr [1:307] "PC1" "PC2" "PC3" "PC4" ...
 - attr(*, "class")= chr "prcomp"
```

17. EDA with PCA

The `prcomp` function also centers the data by default. The centering values are stored in the `center` slot. The `x` slot contains the coordinates of the samples in the new subspace. The

We can plot the samples using the first two PCs as the x and y axes.

```
plot(pca$x[,1], pca$x[,2], pch=20)
```

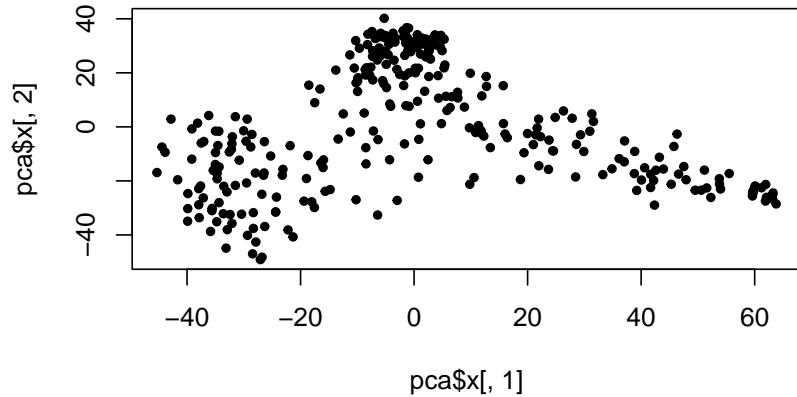


Figure 17.2.: PCA plot of samples in the first two PCs.

If we use ALL the PCs, we can perform a matrix multiplication to get the original data back.

```
orig_data = pca$rotation %*% t(pca$x) + pca$center
orig_data[1:5,1:5]
```

	GSM748053	GSM748054	GSM748055	GSM748056	GSM748057
209125_at	4.400830	4.349534	3.661922	10.289194	3.354648
209988_s_at	3.078285	3.079797	3.467936	3.447669	3.141168
223678_s_at	11.389715	10.637554	5.956832	9.311594	10.467811
218835_at	13.541261	12.944545	9.725079	12.744177	12.896846
201820_at	5.056486	5.030666	4.986433	11.284134	5.132626

Compare to the original data:

```
assay(sub_se, 'exprs')[1:5,1:5]
```

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```
GSM748053 GSM748054 GSM748055 GSM748056 GSM748057  
209125_at     4.400830  4.349534  3.661923 10.289194  3.354648  
209988_s_at   3.078285  3.079797  3.467936  3.447669  3.141168  
223678_s_at   11.389715 10.637554  5.956831  9.311594 10.467811  
218835_at    13.541261 12.944545  9.725079 12.744177 12.896846  
201820_at     5.056486  5.030666  4.986433 11.284134  5.132626
```

And the same thing, but using only the first 3 PCs:

```
orig_data_3pcs = pca$rotation[,1:3] %*% t(pca$x[,1:3]) + pca$center  
orig_data_3pcs[1:5,1:5]
```

```
GSM748053 GSM748054 GSM748055 GSM748056 GSM748057  
209125_at     4.302207  5.319141  4.660544  9.765880  4.368261  
209988_s_at   3.509123  4.372072  4.644271  4.552902  4.251617  
223678_s_at   12.637489 11.381274 10.702921  9.672075 11.985243  
218835_at    14.587942 13.535895 12.807482 12.279706 14.037817  
201820_at     5.300632  6.248694  5.741837 10.170421  5.391310
```

17.5. Variance explained

Often, we want to know how much of the variance in the data is explained by each PC. The `pca` object has a slot called `sdev` that represents the standard deviation of the principle component. Variance is the square of `sdev`, so we can calculate the variance by squaring `sdev`.

```
var_explained = pca$sdev ^ 2
```

The total variance is just the sum of all the variances:

```
tot_variance = sum(var_explained)
```

And the proportion of the variance explained by each PC is then

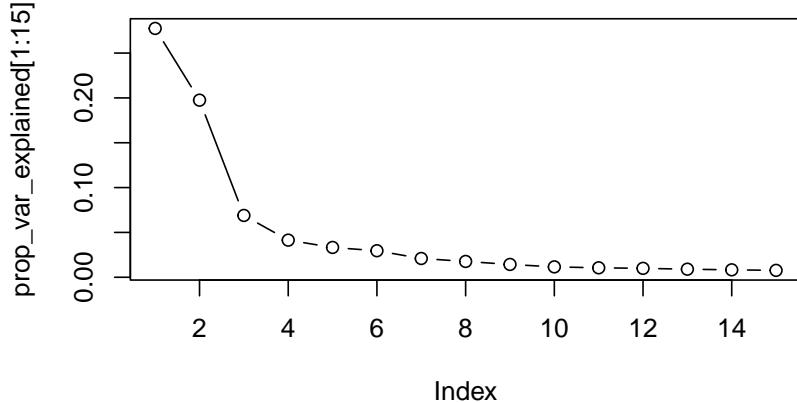
17. EDA with PCA

```
prop_var_explained = var_explained/tot_variance  
head(prop_var_explained)
```

```
[1] 0.27748451 0.19747527 0.06892190 0.04138543 0.03325101 0.02956515
```

If we plot the `prop_var_explained`, it is called a scree plot and can help us to choose an appropriate number of PCs to “keep” in order to reduce the dimensionality.

```
plot(prop_var_explained[1:15], type='b')
```



Examine the plot. How many PCs would you keep?

17.6. Add PCs to our SummarizedExperiment object

Recall that the `x` matrix stored in the `pca` object represent the coordinates of the samples in the new subspace. We can look at the first five rows and columns of the `x` matrix to see what it looks like.

```
pca$x[1:5, 1:5]
```

17. EDA with PCA

	PC1	PC2	PC3	PC4	PC5
GSM748053	0.5712872	34.105929	15.208118	-4.738482	3.4101384
GSM748054	-3.5283223	24.664382	7.632319	-11.746590	0.1405872
GSM748055	5.2892621	21.841834	9.092392	-3.022823	11.7810845
GSM748056	-31.4864198	3.762922	-5.332805	9.695495	-8.8944295
GSM748057	1.2726085	30.640997	10.562888	6.294335	7.1601434

So, PC components for each sample are in columns and samples are in rows. For colData, the samples are also in rows. So, we can join the PC values to the SummarizedExperiment, sub_se, for later use and for comparison to other sample metadata.

```
# We can use cbind to join the PC values to the colData
# note that the names of the rows are the same for both
colData(sub_se) = cbind(colData(sub_se), pca$x)
```

We now have the PCs stored conveniently with our SummarizedExperiment.

```
colnames(colData(sub_se))
```

```
[1] "title"
[2] "geo_accession"
[3] "status"
[4] "submission_date"
[5] "last_update_date"
[6] "type"
[7] "channel_count"
[8] "source_name_ch1"
[9] "organism_ch1"
[10] "characteristics_ch1"
[11] "characteristics_ch1.1"
[12] "characteristics_ch1.2"
[13] "characteristics_ch1.3"
[14] "characteristics_ch1.4"
[15] "characteristics_ch1.5"
[16] "characteristics_ch1.6"
[17] "characteristics_ch1.7"
[18] "characteristics_ch1.8"
```

17. EDA with PCA

```
[19] "characteristics_ch1.9"
[20] "characteristics_ch1.10"
[21] "molecule_ch1"
[22] "extract_protocol_ch1"
[23] "label_ch1"
[24] "label_protocol_ch1"
[25] "taxid_ch1"
[26] "hyb_protocol"
[27] "scan_protocol"
[28] "description"
[29] "data_processing"
[30] "platform_id"
[31] "contact_name"
[32] "contact_laboratory"
[33] "contact_department"
[34] "contact_institute"
[35] "contact_address"
[36] "contact_city"
[37] "contact_zip.postal_code"
[38] "contact_country"
[39] "supplementary_file"
[40] "data_row_count"
[41] "age.at.surgery.ch1"
[42] "disease.free.survival.in.months.ch1"
[43] "follow.up.time..months..ch1"
[44] "gender.ch1"
[45] "histology.ch1"
[46] "pm.stage.ch1"
[47] "pn.stage.ch1"
[48] "pt.stage.ch1"
[49] "relapse..event.1..no.event.0..ch1"
[50] "status.ch1"
[51] "tissue.ch1"
[52] "PC1"
[53] "PC2"
[54] "PC3"
[55] "PC4"
[56] "PC5"
[57] "PC6"
[58] "PC7"
[59] "PC8"
```

17. EDA with PCA

```
[60] "PC9"  
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[64] "PC13"  
[65] "PC14"  
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[100] "PC49"
```

17. EDA with PCA

```
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[103] "PC52"  
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[141] "PC90"
```

17. EDA with PCA

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```

17. EDA with PCA

```
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```

17. EDA with PCA

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```

17. EDA with PCA

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```

17. EDA with PCA

```
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```

17. EDA with PCA

```
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[354] "PC303"  
[355] "PC304"  
[356] "PC305"  
[357] "PC306"  
[358] "PC307"
```

17.7. Variable relationships

Looking at relationships between variables can be a really useful way of generating hypotheses, performing quality control, and suggesting areas to focus in analysis. One common approach to looking at a few variables and their relationships is the “pairs” plot.

The GGally package has a function called ggpairs that can be used to generate a pairs plot for a few variables.

```
library(GGally)
```

Take a look at [this website](#) and examine some variable relationships in the colData(sub_se). When working with ggplot (and ggpairs), you’ll likely want to convert the colData() to a data.frame first. See Figure 17.3 for an example.

```
ggpairs(as.data.frame(colData(sub_se)), columns=(c("PC1", "PC2", "PC3", "gender.ch1")))  
  
'stat_bin()' using `bins = 30`. Pick better value with `binwidth`.  
'stat_bin()' using `bins = 30`. Pick better value with `binwidth`.  
'stat_bin()' using `bins = 30`. Pick better value with `binwidth`.
```

17. EDA with PCA

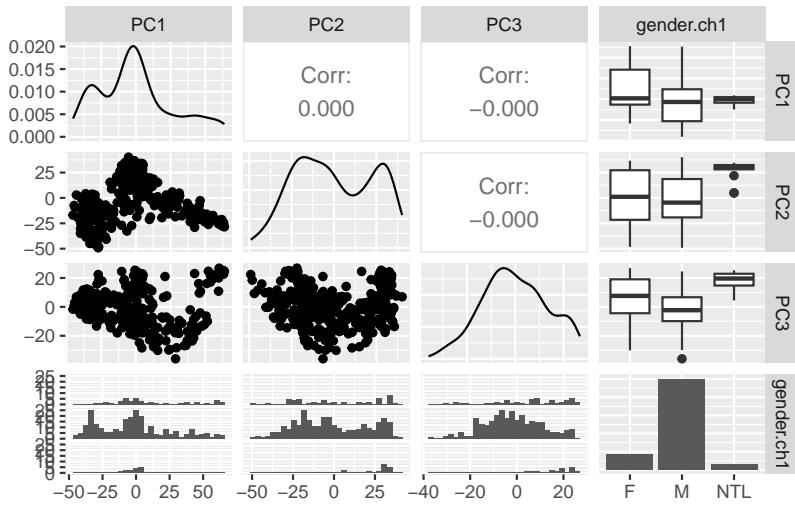


Figure 17.3.: A pairs plot of a few variables.

The ggpairs function is very flexible and plays well with ggplot. Therefore, you can add `aes()` to the `ggpairs` function to add colors, etc. to the plot (see Figure 17.4). Look at other variables that you might want to include and style the plot to your liking.

```
ggpairs(as.data.frame(colData(sub_se)), columns=c("PC1", "PC2", "PC3", "gender.ch1")),
       aes(color=gender.ch1, alpha=0.5))
```

```
`stat_bin()` using `bins = 30`. Pick better value with `binwidth`.
`stat_bin()` using `bins = 30`. Pick better value with `binwidth`.
`stat_bin()` using `bins = 30`. Pick better value with `binwidth`.
```

17. EDA with PCA

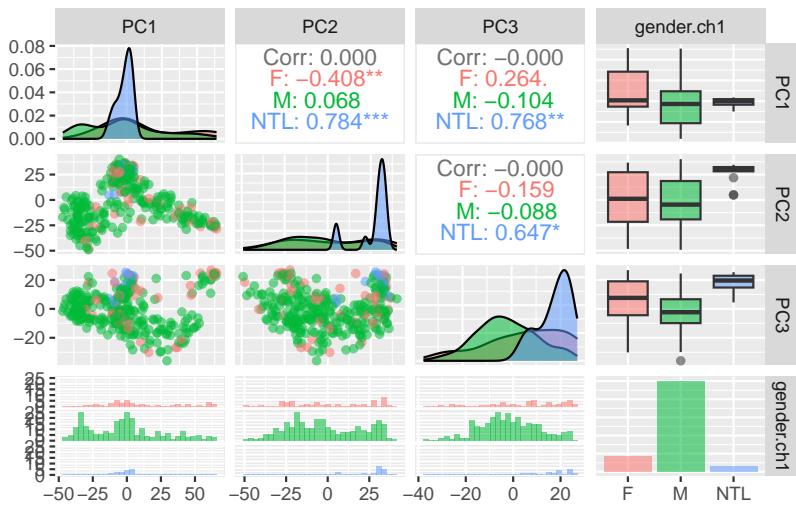


Figure 17.4.: A pairs plot colored by a variable of interest.

References

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A. Appendix

A.1. Data Sets

- [BRFSS subset](#)
- [ALL clinical data](#)
- [ALL expression data](#)

A.2. Swirl

The following is from the [swirl website](#).

The swirl R package makes it fun and easy to learn R programming and data science. If you are new to R, have no fear.

To get started, we need to install a new package into R.

```
install.packages('swirl')
```

Once installed, we want to load it into the R workspace so we can use it.

```
library('swirl')
```

Finally, to get going, start swirl and follow the instructions.

```
swirl()
```

B. Additional resources

- Base R Cheat Sheet

Index

RStudio, [6](#)