BB852 - Data handling, visualisation and statistics

Owen R. Jones

2021-10-08

Contents

Chapter 1

Preface

This book has been written to accompany the course, *BB852 - Data Handling, Visualisation and Statistics*.

It is available as a website (https://jonesor.github.io/BB852_Book/) or as a PDF (click the link at the top of book's website). I recommend to use the website where possible because the formatting is sometimes messy on the PDF, but the PDF is useful if you want a copy for offline use.

> **Note:** The book is a "work in progress" and will change during the course. The latest version can always be found at the website, or by downloading it again. Please let me know (jones@biology.sdu.dk) if you spot any errors, or have any suggestions for improvement.

The book is divided into three parts: data wrangling, data visualisation and statistics.

1.1 Data wrangling

The term data wrangling covers manipulation of data, for example collected from an experiment or observational study, from its raw form to a form that is ready for analysis, or summarised into tables. It includes reshaping, transforming, filtering and augmenting from other data. This book covers these processes in R mainly using the tools from the `dplyr` package.

1.2 Data visualisation

Graphing data is a crucial analytical step that can both highlight problems with the data (e.g. errors and outliers) and can inform on appropriate statistical approaches to take. This book covers the use of `ggplot2` to make high quality, publication-ready plots.

1.3 Statistics

Statistics is a *huge* field and this book does not attempt to cover more than a small fraction of it. Instead it focusses on (ordinary) linear models and generalised linear models. In a nutshell, linear models model the effects of explanatory variables on a continuous response variable with a gaussian (normal) error distribution while generalised linear models (GLMs) offer a more flexible approach that allows the response variable to have non-normal error distributions. This flexibility allows the more-appropriate modelling of phenomena including integer counts (e.g. number of individuals, or species, or events), binary (0/1) data (e.g. survived/died) or binomial count data (e.g. counts of successes and failures). It is important to realise that most commonly-used statistical methods including t-tests, ANOVA, ANCOVA, n-way ANOVA, and of course linear and multiple regression are all special cases of linear models.

My general approach with communicating these methods and ideas is to teach using examples. Therefore, the bulk of the text here consists of walk-throughs of manipulating, plotting and analysing real data. For the statistics section I focus on communicating the "gist" of the underlying mathematical machinery rather than the mathematical details. If you find yourself interested in these details then there are more specialist textbooks available.

This book accompanies the course lectures. The general idea is that there will be a lecture, followed by computer work where you work through the examples in the relevant chapter of this book. At the end of most chapters there are also exercises to test your new skills. It is very important that you do these to gradually build up your skill level and confidence.

1.4 Data sources

This book uses numerous data sets in examples, most of which are real data sets obtained from published works, or collected by me.

The data sets can be found at the following link: https://www.dropbox.com/sh/m8qjm1v7c3bunjg/AACyID24e9VjBflhPzPaN6nza?dl=0

1.5 Your instructors

You are welcome to contact instructors with any problems/questions (but please put a little effort in first).

- Owen Jones, Associate Professor, jones@biology.sdu.dk
- Amandine Aviles, postdoc, amandine@biology.sdu.dk
- Clemens Schauberger, postdoc, schauberger@biology.sdu.dk

1.6 Expectations

There are lectures and practical exercise sessions on the course. The exercise sessions are essential to understand the subject and I expect students to attend and actively participate in them. I also expect students to make every effort to keep up with the core reading (mainly the textbook chapters), and to ask questions where they don't understand. The nature of the course is that it builds sequentially and therefore, if you miss classes or fall behind, it may be hard to catch up. If you don't finish off exercises in class time you should finish them as homework.

1.7 Your feedback

I aim to make this course useful and rewarding for you. I would really like your feedback on how the course is progressing so I can address any issues that come up as soon as possible. To help with this I have created a simple Google Form: https://forms.gle/wZhUfy35ZxEmomYt6. You can use this to send me (Owen) comments (anonymously if you wish) at any time in the course. I promise to do my best to resolve any problems.

1.8 Assessment

The final exam for the course will be a written project where you apply your new skills to analyse some interesting data and report your findings. This will be graded according to the 7-point scale. I will also run some mini-quizzes during the course (e.g. on Kahoot), but these do not count towards your exam.

1.9 Acknowledgements

These materials are inspired by the excellent textbook, "Getting Started With R"[1], which is the recommended textbook for BB852, and by materials for the Sheffield University course "AP 240 - Data Analysis And Statistics With R" (https://dzchilds.github.io/stats-for-bio/). For your convenience, the data sets for the Beckerman et al. book are available at this course's data Dropbox link (above), in a folder called "GSWR_datasets".

[1] Beckerman, Childs & Petchey (2017) *Getting Started With R.* Oxford University Press (2nd edition)

Chapter 2

Schedule

This is the schedule for the course. Please note that it is liable to change (possibly at short notice). If you find a mismatch between this schedule and the official one[1], then it is the official one that is correct.

The columns, GSWR and Course Book, refer to the relevant chapters in the recommended text book ("*Getting Started With R*") and this course book, respectively. You should aim to read and work through these chapters as the course proceeds.

The names of the topics for the **Practical** sessions corresponds to the chapter names in this website/book.

The schedule is only available on the HTML version of this document

[1] https://mitsdu.sdu.dk/skema/activity/N110040101/e21

Chapter 3

Additional recommended reading

These can be downloaded via the link on itsLearning.

- Broman, K. W., & Woo, K. H. (2018). *Data Organization in Spreadsheets.* The American Statistician, 72(1), 2–10.
- Gotelli, N. J., & Ellison, A. M. (2013) Chapter 4, *Framing and Testing Hypotheses*, in A Primer of Ecological Statistics. Sinauer.
- Petchey, O., Beckerman, A., & Childs, D. (2009). *Shock and Awe by Statistical Software - Why R?* Bulletin of the British Ecological Society, 40(4), 55–58.
- Weissgerber, T. L., Milic, N. M., Winham, S. J., & Garovic, V. D. (2015). Beyond bar and line graphs: time for a new data presentation paradigm. PLoS Biology, 13(4), e1002128. doi:%5B10.1371/journal.pbio.1002128](https://doi.org/10.1371/journal.pbio.1002128)
- Wickham, H. (2014). *Tidy Data.* Journal of Statistical Software, 59(10), 1–23.

The following websites are also useful.

The R graph gallery: https://www.r-graph-gallery.com/

STHDA: http://www.sthda.com/english/wiki/ggplot2-essentials http://www.sthda.com/english/wiki/r-basics-quick-and-easy

Chapter 4

An R refresher

In this course we will be learning how manipulate, visualise and analyse data statistically using **R**. **R** is a programming language for data analysis and statistics. It is free and very widely used. One of its strengths is its very wide user base which means that there are hundreds of contributed packages for every conceivable type of analysis. The aim of these introductory sections is to give a basic introduction to the programming language as a tool for importing, manipulating, and exploring data. In later sections we will learn more about statistical analysis.

Before proceeding you will need to ensure you have a recent version of **R** installed on your computer (the version I am using right now is 4.1.0).

Do this: Check your R version, and/or install R on your own computer now.

In this course we will not be using **R** on its own. Instead, we will be using it with RStudio.

R and RStudio are not the same thing. It is possible to run R without RStudio, but RStudio will not work if **R** is not installed. So what is RStudio? RStudio, essentially, is a helpful piece of software that makes **R** easier to use. The three most useful features are:

- The R Console - this is where **R** runs inside RStudio. We can work *directly* with **R** by typing commands into this "console". It is also where outputs (results) from **R** are printed to the screen.
- The Code Editor - this is where you can write **R** programs (called "scripts")", which are a set of commands/instructions in the **R** language saved to a text file. It is much easier to work with scripts using RStudio than with ordinary text editors like Notepad. For example, it colour codes the text to make it easier to read and it will"auto-complete" some text to speed up your work.

- Useful "point-and-click" tools - RStudio can help with tasks like importing data, managing files, reading help files, and managing/installing packages. Doing these things is trickier in *just* **R**: RStudio just makes things easier!

You should do your coding from within RStudio.

You can download **RStudio Desktop** from https://rstudio.com/products/rstudio/download/. Select the correct version for your computer (Mac/Windows) and follow the usual instructions.

Do this: Install **RStudio Desktop** on your computer.

4.1 Getting started with R

In RStudio, create a new "R Script" file. *Scripts* are essentially programs that can be saved to allow you to return to your work in the future. They also make debugging of errors much easier.

You can use the menu to do create a new R Script (`File > New File > R Script`), but there's also a keyboard shortcut (Windows: `Ctrl+Shift+N`; Mac: `Cmd+Shift+N`). If you save (Windows: `Ctrl+S`; Mac: `Cmd+S`), you will be prompted for a file name. Make sure it has the suffix ".R" which denotes an R script file. Save the file in a folder with a memorable name (e.g. `BB852_Work`).

When you double click on this file in future, it should automatically open in RStudio (if it doesn't you should be able to right-click and select `Open with...`).

In RStudio you can execute commands using the "run" icon at the top of the script window, or by selecting the text and typing the shortcut `Ctrl+Enter` (Windows) or `Cmd+Enter` (Mac). Another helpful feature of RStudio is that it will colour-code the syntax that you type, making it easier to read and debug. Note that the colours you see may be different from the ones shown in this handout.

You can customise the look of RStudio using by clicking **Tools → Options** menu on Windows or **RStudio → Preferences** on a Mac. I will point out some of this in the lecture, or you can ask me to show you.

Over the next few pages I will introduce the basics of the R programming language. Try typing them into the scripting window (top left) in RStudio and ensuring that you understand what the commands are doing. It is impossible to "break" R by typing the wrong command so I encourage you to experiment and explore the R language I introduce to you here as much as possible - it really is the best way to learn!

The "look" of RStudio can be modified by changing the Preferences (**RStudio → Preferences → Appearance**). Also, there are some useful keyboard shortcuts that are worth learning, to run code, save files etc. without needing to point-and-click (**Tools → Keyboard Shortcuts Help**).

4.2 Getting help

R features a wealth of commands, which are more properly termed **functions**. You will learn many of these over the next few weeks. Functions often feature a several options which are specified with **arguments**. For example, the function `sum`, has the argument `...`, which is intended to be one or more **vectors** of numbers (see below), and the argument `na.rm`, which is a logical argument specifying whether or not missing values should be removed or not. Usually the arguments have default options which are used you choose not to specify them. In addition, you don't necessarily need to fully-specify the argument if they are specified in the *correct order*.

You can get **help** on R functions from within R/RStudio with the `?` and `help.search` commands. `?` requires that you know the function name while *help.search* will search all the available help files for a particular word or phrase. `??` is a synonym for *help.search*:

```
?rep
help.search("bar plot")
??"bar plot"
```

In RStudio, the help results will appear in the lower right hand area.

4.3 R as a fancy calculator

R features the usual arithmetic operations for addition, subtraction, division, multiplication:

```
4 + 3
```

```
## [1] 7
```

```
9 - 12
```

```
## [1] -3
```

```
6 / 3
```

```
## [1] 2
```

```
7 * 3
```

```
## [1] 21
```

```
(2 * 7) + 2 - 0.4
```

```
## [1] 15.6
```

R also has commands for square root (`sqrt`), raising to powers (`^`), taking the absolute value (`abs`), and rounding (`round`), natural log (`log`), anti-log (`exp`), log to base-10 (`log10`):

```
sqrt(945)
```

```
## [1] 30.74085
```

```
3^5
```

```
## [1] 243
```

```
abs(-23.4)
```

```
## [1] 23.4
```

```
round(2.35425, digits = 2)
```

```
## [1] 2.35
```

```
log(1.2)
```

```
## [1] 0.1823216
```

```
exp(1)
```

```
## [1] 2.718282
```

```
log10(6)
```

```
## [1] 0.7781513
```

Another thing you can do is evaluate TRUE/FALSE conditions:

```
3 < 10
```

```
## [1] TRUE
```

```
5 > 7
```

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

```
5 == 5
```

```
## [1] TRUE
```

```
6 != 5
```

```
## [1] TRUE
```

```
3 %in% c(1, 2, 3, 4, 5)
```

```
## [1] TRUE
```

```
6 %in% c(1, 2, 3, 4, 5)
```

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

4.4 Objects in R

R is an object oriented programming language. This means that it represents concepts as **objects** that have data fields describing the object. These objects can be manipulated by **functions**. Objects can include data, but also models. Don't worry about these distinctions too much for now - all will become clear as you proceed!

Objects are assigned names in R like this. The "`<-`" command is pronounced "*gets*" so I would pronounce the following as "*x gets four*":

```
x <- 4
```

To look at any object (function or data), just type its name.

```
x
```

```
## [1] 4
```

The main data object types in R are: *vectors*, *data frames*, *lists* and *matrices*. We will focus on the first two of these during this course.

A vector is simply a series of data (e.g. the sequence *1, 2, 3, 4, 5* is a vector, so is the non-numeric sequence *Male, Female, Female, Male, Male*). Each item in a vector is called an **element**. Therefore, both of these examples contain 5 elements.

There are several ways to create vectors in R. For example, you can make vectors of integers using the *colon* (`:`) function (e.g. `1:5`), or vectors of any kind of variable using the `c` function. `c` stands for *concatenate*, which means to *join (things) together in a chain or series*. Other convenient functions for making vectors are `seq`, which builds a sequence of numbers according to some rules, and `rep` which builds a vector by repeating elements a specified number of times.

Try the following:

```
A <- 1:5
B <- c(1, 3, 6, 1, 7, 9)
C <- seq(1, 12, 2)
D <- seq(1, 5, 0.1)
E <- rep(c("Male", "Female"), each = 3)
G <- rep(c("Male", "Female"), c(2, 4))
```

Try modifying the commands to make sure you know what the commands are doing.

Chapter 5

Manipulating objects

Objects can be manipulated (just like in real life). In R, we use **functions** to manipulate objects.

For example, we can use the basic arithmetic functions (`*`, `+`, `/`,`-`) on a vector:

```
B
```

```
## [1] 1 3 6 1 7 9
```

```
B * 3
```

```
## [1]  3  9 18  3 21 27
```

```
B - 2
```

```
## [1] -1  1  4 -1  5  7
```

You can *concatenate* entire vectors together using the `c` function. E.g. concatenating the vectors `A` and `B` from above:

```
c(A, B)
```

```
##  [1] 1 2 3 4 5 1 3 6 1 7 9
```

Other manipulations are also done "element-by-element". For example, here we multiply the first element of B by 1, the second by 2, the 3rd by 3 and so on…:

```
B * c(1, 2, 3, 4, 5, 6)
```

```
## [1]  1  6 18  4 35 54
```

If the length of the vectors match, we can also multiply (or add/subtract/divide etc.) multiple vectors:

```
A / B
```

```
## [1] 1.0000000 0.6666667 0.5000000 4.0000000 0.7142857 0.1111111
```

5.1 Missing values, infinity and "non-numbers"

By convention, *missing values* in R are coded by the value "NA". The way that particular functions handle missing values varies: sometimes the NA values are stripped out of the data, other times the function may fail.

For example, if we asked for the mean value of a vector of numbers with an NA value, it will fail:

```
mean(c(1, 3, 6, 1, 7, 9, NA))
```

```
## [1] NA
```

In this case you need to specify that any NA values should be removed before calculating the mean:

```
mean(c(1, 3, 6, 1, 7, 9, NA), na.rm = TRUE)
```

```
## [1] 4.5
```

Calculations can sometimes lead to answers that are plus, or minus, infinity. These values are represented in R by `Inf` or `-Inf`:

```
5 / 0
```

```
## [1] Inf
```

```
-4 / 0
```

```
## [1] -Inf
```

Other calculations lead to answers that are not numbers, and these are represented by `NaN` in R:

```
0 / 0
```

```
## [1] NaN
```

```
Inf - Inf
```

```
## [1] NaN
```

5.2 Basic information about objects

You can obtain information about most objects using the `summary` function:

```
summary(B)
```

```
##    Min. 1st Qu.  Median    Mean 3rd Qu.    Max.
##    1.00    1.50    4.50    4.50    6.75    9.00
```

The functions `max`, `min`, `range`, and `length` are also useful:

```
max(B)
```

```
## [1] 9
```

```
min(B)
```

```
## [1] 1
```

```
range(B)
```

```
## [1] 1 9
```

```
length(B)
```

```
## [1] 6
```

5.3 Data frames

Data frames are the usual way of storing data in R. It is more-or-less the same as a worksheet in Excel. A data frame is usually made up of a number of vectors (of the same length) bound together in a single object. You can make a data frame by binding together vectors, or you can import them from outside R.

This example shows the creation of a data frame in R, from 3 vectors:

```
height <- c(173, 145, 187, 155, 179, 133)
sex <- c("Male", "Female", "Male", "Female", "Male", "Female")
age <- c(17, 22, 32, 20, 27, 30)

mydata <- data.frame(height = height, age = age, sex = sex)
mydata
```

```
##   height age    sex
## 1    173  17   Male
## 2    145  22 Female
## 3    187  32   Male
## 4    155  20 Female
## 5    179  27   Male
## 6    133  30 Female
```

Data frames can be summarised using the `summary` function (or the `str` function, which gives you a different view of the same data):

```
summary(mydata)
```

```
##      height           age           sex
##  Min.   :133.0   Min.   :17.00   Length:6
##  1st Qu.:147.5   1st Qu.:20.50   Class :character
##  Median :164.0   Median :24.50   Mode  :character
##  Mean   :162.0   Mean   :24.67
##  3rd Qu.:177.5   3rd Qu.:29.25
##  Max.   :187.0   Max.   :32.00
```

```
str(mydata)
```

```
## 'data.frame':    6 obs. of  3 variables:
##  $ height: num  173 145 187 155 179 133
##  $ age   : num  17 22 32 20 27 30
##  $ sex   : chr  "Male" "Female" "Male" "Female" ...
```

Data frames can be subsetted using the square brackets `[]`, or `subset` functions. With the square brackets, the first number specifies the row number, while the second number specifies the column number:

```
mydata[1, ]
```

```
##   height age  sex
## 1    173  17 Male
```

```
mydata[, 2]
```

```
## [1] 17 22 32 20 27 30
```

```
mydata[1, 2]
```

```
## [1] 17
```

```
subset(mydata, sex == "Female")
```

```
##   height age    sex
## 2    145  22 Female
## 4    155  20 Female
## 6    133  30 Female
```

5.4 Classes in R

Every objects you create, or import into R, has a "type" called a `class`. You can ask what class an object has using the `class` function.

For example, the vectors you created above have types.

```
class(height)
```

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

```
class(sex)
```

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

```
class(mydata)
```

```
## [1] "data.frame"
```

You can find out the class of all columns in a `data.frame` by asking for a summary with `str`. For example, in this example, there are two numeric columns (`num`) and a character column (`chr`).

```
str(mydata)
```

```
## 'data.frame':    6 obs. of  3 variables:
##  $ height: num  173 145 187 155 179 133
##  $ age   : num  17 22 32 20 27 30
##  $ sex   : chr  "Male" "Female" "Male" "Female" ...
```

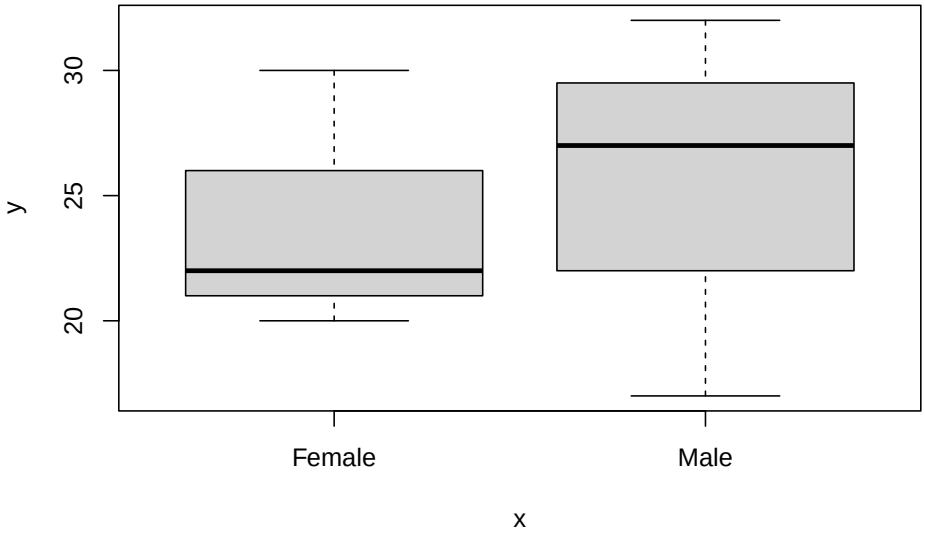
There's another special class of vector called `factor`. In the small dataset above (`mydata`), sex is registered by R to be a `character` vector. For some functionality this is perfectly fine, but for others you will need to convert the data into a factor.

For example, this code, to make a box plot, will not work:

```
plot(mydata$sex, mydata$age)
```

But this code will work fine:

```
plot(as.factor(mydata$sex), mydata$age)
```

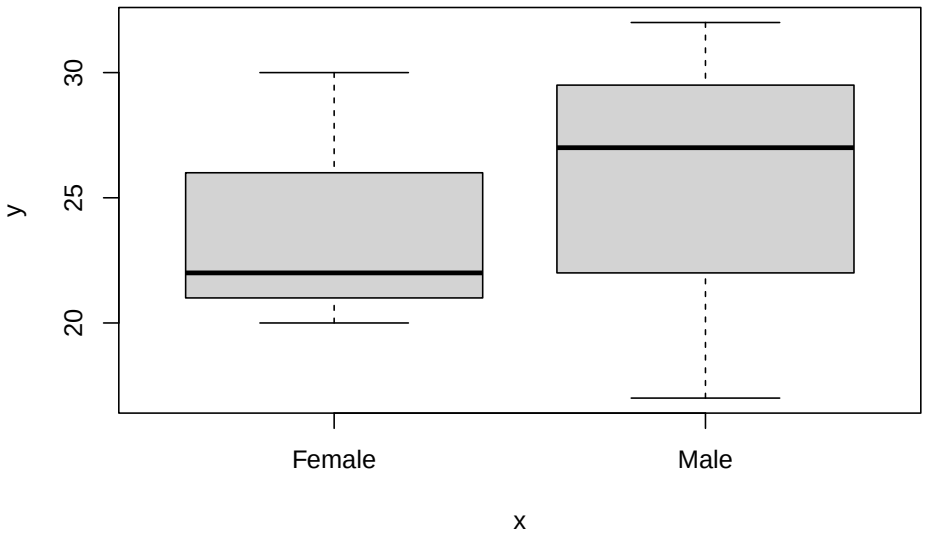


Of course it might be easier to convert it to be a factor in the data frame itself, like this:

```
mydata$sex <- as.factor(mydata$sex)
str(mydata) # You can see that it is now registered as a factor
```

```
## 'data.frame':    6 obs. of  3 variables:
##  $ height: num  173 145 187 155 179 133
##  $ age   : num  17 22 32 20 27 30
##  $ sex   : Factor w/ 2 levels "Female","Male": 2 1 2 1 2 1
```

```
plot(mydata$sex, mydata$age)
```



If you are getting strange results from your code it is a good idea to check the structure of the data. Are the classes what they should be?

5.5 Organising your work

It would be incredibly tedious to enter real data into **R** by typing it in!

Thankfully, R can import data from a several data formats, and it understands the file structure of your computer. Thus, you can use spreadsheet software (like Excel) to enter and store your data, and you can organise your project work in a sensible way in folders (sometimes called *directories*) on your computer.

The most commonly used data format is `comma separated value (CSV)` so I will use that. You can also import from Excel, but the data must be formatted in a particular way to enable this (I'll cover this in a later class).

For this course, I suggest that you make a folder somewhere on your computer called "IntroToR". We will use this as the `working directory` for the remainder of the session. In RStudio you can set the working directory by clicking through the menu items **Session → Set Working Directory → Choose Directory**.

You can also using the `setwd` function to do this, if you know where your files are stored (the *file path*). File paths in Windows and Mac computers are expressed differently. Apple systems use the forward-slash (/) to separate folders whereas Windows can use the forward-slash (/) or double-backslash (\). In windows you also need to define the drive (e.g. C:).

So, to set the working directory in Apple OSX you would use something like this (obviously, you need to put *your* path!):

```
setwd("/Users/orj/Desktop/IntroToR")
```

While in Windows the equivalent command would be something like this (both of the following should work):

```
setwd("C:\\Users\\orj\\Desktop\\IntroToR")
setwd("C:/Users/orj/Desktop/IntroToR")
```

Typing the path in can be annoying but there are ways to speed it up. In Windows you can copy paths from the Windows Explorer location/address bar, or you can hold down the Shift key as you right-click the file, and then choose Copy As Path.

On a Mac you can copy file paths from Finder: Select your file/folder, Right click, Press the option key (on my keyboard this is the `alt` key) and click "Copy X as Pathname"

I can check what the current working directory is using the `getwd` function:

```
getwd()
```

It is good practice to keep your files well-organised. I recommend that you create a folder in your working directory called `CourseData` (or similar). Store your data files in this folder.

I have put all the data for the course into a Dropbox folder - see the link in Chapter 1. In there you will find a file called "`carnivora.csv`". Download this to your new `CourseData` folder.

You can now import this file into R using the `read.csv` function. The specification of the argument `header = TRUE` signifies that the first row of our CSV file contains the column names. Note that your file path will be different to mine[1]:

```
carni <- read.csv("CourseData/carnivora.csv",
  header = TRUE,
  stringsAsFactors = TRUE
)
```

The `stringsAsFactors` argument tells R to treat text-type data (technically known as "character **strings**") as a special kind of data called **factors**. Essentially, factors are *categorical* data where the data can take a limited number of discrete values. For example, "treatmentA", "treatmentB", "treatmentC". Although this may seem a little esoteric right now, it is important to ensure that your data is recognised by R in the correct way. In **most** cases, your text-type data will be factor data, so it is usually safe to set `stringsAsFactors = TRUE`.

> **Tip:** RStudio also has a point-and-click "Wizard" to help import data. Look for "Import Dataset" in the top-right pane.

5.6 Inspecting the data

We can get some basic information on your imported data (e.g. the `carni` data frame) using the `summary` function, but also the `dim` and `nrow/ncol` functions:

```
summary(carni)
```

```
dim(carni)
```

```
## [1] 112  17
```

[1] A note here about code formatting: You can see that I have written the code over several lines. This is not strictly necessary, but (I think) it can make long commands easier to read. R doesn't "see" the new lines. The plot command could be in a single long line.

```
nrow(carni)
```

```
## [1] 112
```

```
ncol(carni)
```

```
## [1] 17
```

We can find the names of the columns of a data frame with the `names` function:

```
names(carni)
```

```
##  [1] "Order"       "SuperFamily" "Family"      "Genus"       "Species"
##  [6] "FW"          "SW"          "FB"          "SB"          "LS"
## [11] "GL"          "BW"          "WA"          "AI"          "LY"
## [16] "AM"          "IB"
```

The first few columns are to do with the taxonomic placement of the species (Order, SuperFamily, Family, Genus and Species). There then follow several columns of life history variables: FW = Female body weight (kg), SW = Average body weight of adult male and adult female (kg), FB = Female brain weight (g), SB = Average brain weight of adult male and adult female (g), LS = Litter size, GL = Gestation length (days), BW = Birth weight (g), WA = Weaning age (days), AI = Age of independence (days), LY = Longevity (months), AM = Age of sexual maturity (days), IB = Inter-birth interval (months).

You can refer to the sub-parts of a `data.frame` (the columns) using the `$` syntax:

```
summary(carni$FW)
```

```
##    Min. 1st Qu.  Median    Mean 3rd Qu.    Max.
##   0.050   1.245   3.400  18.099  10.363 320.000
```

5.7 "Classes" in R

I have already mentioned the different object types in R (e.g. vectors and data frames). The object types are technically known as "classes". You can find out what "class" an object is by using the `class` function:

```
class(carni)
```

```
## [1] "data.frame"
```

In this case, the data frame is, unsurprisingly, of class "data.frame". However, the vectors that compose the data frame also have classes. There are several classes of vectors including "integer" (whole numbers), "numeric" (real numbers), "factor" (categorical variables) and "logical" (true/false values).

I expect you have heard of the first two data types, but "factor" might be puzzling. Factors are defined as variables which can take on a *limited* number of different values. They are often referred to as `categorical variables`. For example, in the carnivore dataset, the taxonomic variables are factors. The different values that a factor can take are known as `levels` and you can check on the levels of a vector with the `levels` function.

```
class(carni$Family)
```

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

```
levels(carni$Family)
```

```
## [1] "Ailuridae"   "Canidae"     "Felidae"     "Hyaenidae"   "Mustelidae"
## [6] "Procyonidae" "Ursidae"     "Viverridae"
```

5.8 Tables and summary statistics

For vectors of class "factor" you can use the `table` function to give the counts for each level:

```
table(carni$Family)
```

```
##
##   Ailuridae     Canidae     Felidae   Hyaenidae  Mustelidae Procyonidae
##           1          18          19           4          30           4
##     Ursidae  Viverridae
##           4          32
```

You can use the function `tapply` ("table apply"), to get more complex summary information. For example, I could ask what the mean female weight (FW) is in each of the families using the argument `mean`:

```
tapply(carni$FW, carni$Family, mean)
```

```
##   Ailuridae     Canidae     Felidae   Hyaenidae  Mustelidae Procyonidae
##  120.000000    9.050000   31.432105   33.540000    3.989000    3.642500
##     Ursidae  Viverridae
##  198.250000    2.672813
```

5.9 Plotting data

Basic plots can be made using the `plot` command. For example, let's have a look at the relationship between log gestation length and log female body weight (see Figure 5.1, below):

```
plot(log(carni$FW), log(carni$GL))
```

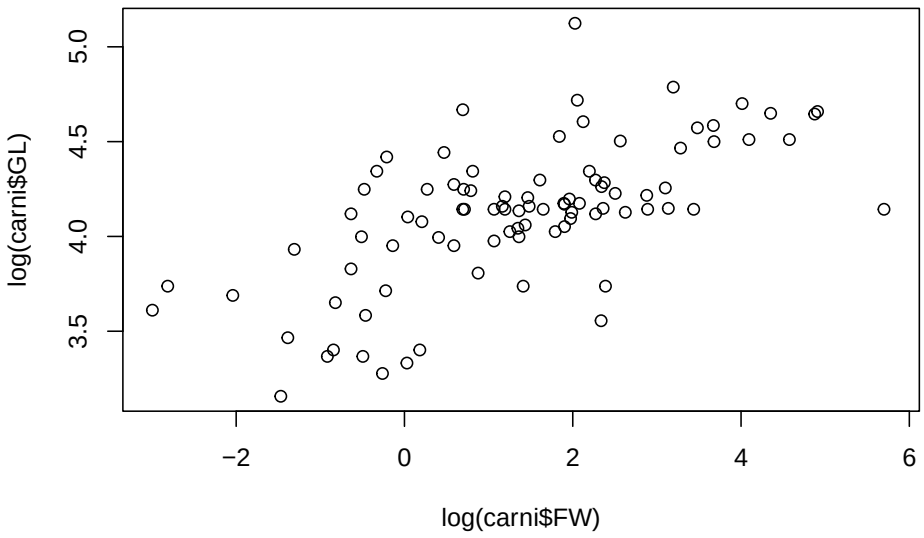


Figure 5.1: A simple scatter plot

5.10 R Packages

R packages are collections of software that add capabilities to "base R". In this course we use several packages including `dplyr`, which adds functionality for manipulating data, `ggplot2` which helps us make pretty plots and `magrittr` which adds tools to allow more "elegant" programming. Packages need to be installed using `install.packages` command before they can be used. You only need to install them once.

```
install.packages("dplyr")
install.packages("ggplot2")
install.packages("magrittr")
```

To use the packages you need to load them with the `library` command, like this:

```
library(dplyr)
library(ggplot2)
library(magrittr)
```

We will be using these packages a lot, and you will need to remember to load them every session. It is therefore useful to add those `library` commands to the top of every script you write.

5.11 Exercise: Californian bird diversity

In the 1950s-1970s there was rapid growth in the number of houses being built in California, with suburbs sprawling out into the new sites in the countryside. What effect would this have on local bird communities?

Surveys on bird abundances were carried out in several locations near Oakland, California [2]. The locations were of different ages, enabling us to investigate what changes might happen through time. Although there were no surveys before the developments, we can regard the bird abundance in the very youngest housing developments as the baseline pre-development condition.

Think about what you might expect to happen to bird species diversity through time in a newly developing suburb.

5.11.1 The data

The relevant data file is called `suburbanBirds.csv`. This file contains data on bird abundances surveyed in 1975. The columns of the data are `Name` (name of the suburb), `Year` (the year that the suburb was built), `HabitatIndex` (an index of habitat quality, related to tree height, garden maturity etc.), `nIndividuals` (number of individual birds seen in a standard survey) and `nSpecies` (number of species seen in a standard survey).

Additional surveys found an average species richness of 3.5 in nearby undisturbed habitats of grassland savanna.

5.11.2 Try the following

1. First import the data. Check that the columns look as they should (use `summary` or `str` functions).
2. What is the mean, minimum, and maximum number of species seen? (there is more than one way to do this)
3. How old are the youngest and oldest suburbs? (hint: the survey was carried out in 1975, do the maths!)

[2] Vale, T. R., & Vale, G. R. (1976). Suburban bird populations in west-central California. Journal of Biogeography, 157–165.

4. Plot the relationship between `Year` and `nSpecies` as a scatter plot using base-R graphics (using the `plot` function).

5. The pattern might be easier to see if you could replace `YearBuilt` with suburb age. Create a new vector in your data frame for this variable (e.g. `df$Age <- 1975 - Year`). Re-plot your results.

6. What do the data show? What might be the mechanisms for the patterns you see? Do they match your expectations?

7. Export your plots and paste them into a Word Document.

8. If you get this far, try plotting the other variables in the dataset.

Chapter 6

Tips and tricks

6.1 Appearance

You can modify the appearance of RStudio via the *Options* dialog: **Tools > Options** menu (**RStudio > Preferences** on a Mac).

There you will find many ways of modifying how RStudio looks and works. However, for beginners I suggest to leave the defaults for most of these. Instead, focus on the Appearance section where you can change the colour scheme or "Theme". My current favourite is "Modern", what's yours? You can also change the font and font size. When choosing a font, it is important to use only "monospace" fonts (these are fonts fixed width of characters). Why would you want to do that? Some fonts are better at distinguishing similar looking characters (e.g. is that an upper case or lower case letter "w", or is it a "1" (the number one) or a "l" (lower case L)). Good examples of monospace fonts are "LucidaConsole", and "Courier". The availability of fonts might differ between computers.

If you choose non-monospaced fonts it can cause problems with formatting your code in the scripts, so if you get any formatting weirdness, check your font!

6.2 Shortcuts

There are several useful keyboard shortcuts that can make your life easier in RStudio. You can find a full list by clicking through the menu – Tools > Keyboard Shortcuts Help – but these are the ones I find most useful:

- Auto-complete code: if you start to write function or object names, after a short pause, RStudio will offer some auto-complete the name. Use the arrow keys to choose the best option, and press Enter.
- Run current line/selection Ctrl+Enter (Cmd+Return, on a Mac)
- Run code from script beginning to current line: Ctrl+Alt+B (Cmd+Option+B)

- Comment/uncomment current line/selection: Ctrl+Shift+C (Cmd+Shift+C)
- Reflow Comment: Ctrl+Shift+/ (Cmd+Shift+/)
- Find text in Files: Ctrl+Shift+F (Cmd+Shift+F)
- Undo: Ctrl+Z (Cmd+Z)
- Cut: Ctrl+X (Cmd+X)
- Copy: Ctrl+C (Cmd+C)
- Paste: Ctrl+V (Cmd+V)
- Parentheses (brackets): Select text you want to "wrap" in parentheses and type Shift+(, or Shift+), to automatically put the brackets on both sides of text.
- Pipes (`%>%`: Ctrl+Shift+M (Cmd+Shift+M) (you'll meet these later in the course!)

6.3 Code style

R is very forgiving about code styling, the use of white space (e.g., spaces and tabs), splitting code over lines and so on. This can make things a bit messy sometimes so it is a good idea to develop a style that you find easy to work with.

There are some guidelines for "good style" which you might like to follow e.g. https://google.github.io/styleguide/Rguide.html or https://style.tidyverse.org.

There is also a way to automatically format your code into a consistent, nice, style after you have written it. To do that, you use the `styler` R package, and then add a keyboard/menu shortcut to RStudio (click Tools > Addins). See here: https://lorenzwalthert.github.io/stylerpost/.

6.4 The plots pane

As you make plots they appear in the plot pane, which is by default on the bottom right of RStudio. Old plots are kept in the memory and you can navigate to them using the back/forward arrows at the top of the plot pane.

You can use the *Export* button to copy a plot (for pasting into a document) after resizing it, or to save a plot as an image file.

6.5 Tables

If you have made summary tables, for example with the `dplyr` function `summarise`, you can save them as a csv file using `write.csv(xxx, file = "xxx.csv", row.names = FALSE)`. You can then open the csv in Excel and cut/paste into your Word document as a table.

6.6 Importing data from text files

There are several ways of importing data, and several ways that it can go wrong.

RStudio can import data in text files via the *Import Dataset* button (top right pane), or via the the main menu (File > Import Dataset). There are two ways to import data from text files (such as .csv or .tab or .txt). These are `base` and `readr`. They work in similar ways. During the import process you can choose what the column delimiter (separator) is (e.g. `,` or `;`) and what the decimal separator is (e.g. `.` or `,`).

When you have imported the data, you should check it using e.g. `head` or `str` functions. Make sure that you have the expected number of columns, and that they have the right names. Most of the problems with importing data comes from problems with Excel. A common problem is that the columns of the data are squashed together into a single column. This can be because (1) you have chosen the wrong delimiter (separator) or (2) because Excel has saved the csv file incorrectly.

To avoid this problems (1) check the delimiter (2) avoiding saving the file with Excel when you download it. You can also open the text file in a text editor and remove problematic characters using "Find and Replace". For example, the single-column problem is caused by Excel putting the line of data within quotation marks, which you could remove and then save the data.

6.7 Importing data from Excel

Yes, it is possible to import data directly from Excel using the *Import Dataset* dialogue (or using the `read_excel` function from the `readxl` package). This is pretty neat – BUT – the data need to be very well-arranged for this to work properly. What do I mean by "well-arranged"? Read the paper by Broman & Wu for more details [1]

6.8 Numbers

R uses scientific notation when showing you numbers. Thus very large and very small numbers are shown with exponentials so that 1,000,000 is shown as `1e+06` and `0.0005` is shown as `5e-04`, for example. This can be a bit confusing, and you can turn this behaviour off by setting the `scipen` option like this.

```
options(scipen = 999)
```

The option will be set until you re-start R, or until you turn it off with `options(scipen = 0)`. You could start your script by setting this option.

[1] Broman, K. W., & Woo, K. H. (2018). *Data Organization in Spreadsheets.* The American Statistician, 72(1), 2–10.

Chapter 7

Paths and projects

7.1 File structure

Files on a computer are stored in hierarchical folders (also known as directories). On a Mac you can use the Finder program to navigate these folders while on a Windows machine you can use Windows Explorer.

Try that now!

When managing your research projects, it's a good idea to keep your files organised in some sensible structure rather than simply dumping all your files chaotically into the same folder. Be kind to your future self and get organised!

Exactly how you do that is a matter of personal taste but my own personal preference is to set up a folder per project, and then, within that folder to create folders for Data, Code (for my R scripts), Plots (graphs produced by R), and Writing (e.g. for Word documents with my project write-up).

7.2 Paths and the command line

The Command Line Interface (CLI) is an program interface where you use text commands to operate the program rather than the now more commonplace Graphical User Interface (GUI). R and Rstudio have a mix of CLI and GUI.

To use files (e.g. data) in CLI you will need to know the path, which can be written in text with folder names separated by slashes

e.g. .`C:/Users/Owen/Documents/ProjectX/Data/myData.txt`

7.3 R and file structure

To load data into R using the CLI you need to use file paths which can be annoying to type.

e.g. `x <- read.csv("C:/Users/Owen/Documents/Analysis/SurveyAnalysis1/Data/myData.txt")`

There are two ways to make life easier for yourself: (1) you can set the "working directory" for your project; (2) you can set up an R Project.

It is true that RStudio has a data import wizard to help with this, but setting a working directory or using Projects is recommended. I will briefly outline these two options.

7.3.1 Setting the working directory

R/RStudio use "relative paths" which means that you can tell R where you are working (i.e. the path). To understand what this means, is is useful to see what R sees:

Open Rstudio and type `getwd()` ("get working directory") to get the working directory of your RStudio session. Any files within that working directory folder can be loaded **without** typing the full path. In other words, if a data file, `myData.csv`, is in the working directory folder you could load it by typing `x <- read.csv("myData.txt")`rather than with the full path e.g. `x <- read.csv("C:/Users/Owen/Documents/Analysis/SurveyAnalysis1/Data/myData.csv")`.

In R you can change the working directory with `setwd()` e.g. `setwd("PATH_TO_NEW_DIRECTORY")`.

Basically, setting the working directory acts like a short cut. By setting the working directory to like this:

`setwd("C:/Users/Owen/Documents/Analysis/SurveyAnalysis1/")`,

the long command to read in data…

`x <- read.csv("C:/Users/Owen/Documents/Analysis/SurveyAnalysis1/Data/myData.csv")`

…becomes much shorter…

`x <- read.csv("Data/myData.txt")`.

7.4 Projects in R

Projects in RStudio are a very convenient way to automate the setting of the working directory. To set up a project do the following:

- Navigate in *Finder* (Mac) or *Windows Explorer* (Windows) to where you would like to put your work.
- Create a folder to contain your work (e.g. `BB852CourseWork`, this is your Working Directory.
- Open RStudio.
- Click `File > New Project`.
- Click `Existing directory`.
- Browse to find the correct folder, click to enter the folder.

- Click `Create Project`. This will create a file called e.g. `BB852CourseWork.Rproj`. From now on, you can open RStudio by clicking this file. Doing so will automatically set your working directory and any other settings saved for your project.
- Close RStudio and try opening the `Rproj` file by clicking it.

If you use the command `getwd()` you will see that the working directory is now automatically set as the project folder location. Awesome!

After you have created a project folder (e.g. `BB852CourseWork`), you can add useful folders to keep yourself organised. As mentioned above, exactly how you do that is a matter of personal taste but you should at least have a folder here for `data` and `plots`.

Part I

Data Wrangling

Chapter 8

Data wrangling with `dplyr`

This chapter focuses on using the package `dplyr`, which is designed to make working with data in R easier. The package has several key "workhorse" functions, sometimes called **verbs**. These are: `filter`, `select`, `mutate`, `arrange` and `summarise`. I covered these in the lecture, and they are also discussed in the textbook. This chapter guides you through worked examples to illustrate their use.

We will also be using **pipes** from the `magrittr` package. These are implemented using the command `%>%`.

```
library("dplyr")
library("magrittr")
```

To get to know `dplyr` and its functions we'll use a data set collected from the university campus at University of Southern Denmark (SDU) The SDU bird project follows the fate of mainly great tits (musvit) and blue tits (blåmejse) in about 100 nest boxes in the woods around the main SDU campus.

We will address two questions concerning clutch size (the number of eggs laid into the nest) -

1. How does clutch size differ between blue tits and great tits?
2. How does average clutch size vary among years?

To answer these questions we need to calculate the average clutch size (number of eggs) for each nest in each year. The data are in a file called (`sduBirds.csv`) and are raw data collected while visiting the nests. The data will need to be processed to answer those questions.

Let's import the data and take a look at it. Make sure your data looks OK before moving on. You should first set up your working directory (e.g. a folder for the course, with a sub-folder for course data etc.), and set it (with `setwd`)). See the earlier material for how to do this, or ask for help.

```
df <- read.csv("CourseData/sduBirds.csv")
str(df)
```

```
## 'data.frame':    9357 obs. of  15 variables:
##  $ Timestamp  : chr  "2013-05-14" "2013-05-03" "2013-06-25" "2013-06-18" ...
##  $ Year       : int  2013 2013 2013 2013 2013 2013 2013 2013 2013 2013 ...
##  $ Day        : int  134 123 176 169 112 112 116 183 143 107 ...
##  $ boxNumber  : int  1 1 1 1 1 1 1 1 1 1 ...
##  $ species    : chr  "BT" "BT" "BT" "BT" ...
##  $ stage      : chr  "NL" "NL" "NE" "NE" ...
##  $ nEggs      : int  12 8 0 0 0 0 0 0 NA 0 ...
##  $ nLiveChicks: int  0 0 0 0 0 0 0 0 0 0 ...
##  $ nDeadChicks: int  0 0 0 0 0 0 0 0 0 0 ...
##  $ eggStatus  : chr  "WA" "CO, CV" NA NA ...
##  $ chickStatus: chr  NA NA NA NA ...
##  $ adultStatus: chr  "FN" "FN" NA NA ...
##  $ finalStatus: chr  NA NA "NE" "NE" ...
##  $ Comments   : chr  NA "MA" NA NA ...
##  $ observerID : chr  "AMK" "AMK" "AMK" "AMK" ...
```

8.1 `select`

From the `str` summary (above) you can see that there are many columns in the data set and we only need some of them. Let's `select` only the columns that we need for our calculations to make things a bit easier to handle. We need the `species`, `Year`, `Day`, `boxNumber` and `nEggs`:

```
df <- select(df, species, Year, Day, boxNumber, nEggs)
head(df)
```

```
##   species Year Day boxNumber nEggs
## 1      BT 2013 134         1    12
## 2      BT 2013 123         1     8
## 3      BT 2013 176         1     0
## 4      BT 2013 169         1     0
## 5      BT 2013 112         1     0
## 6      BT 2013 112         1     0
```

The output of `head` shows you the first few rows of the data set. You can see that each row represents a visit of a researcher to a particular nest. The researcher records the bird species if it is known (GT = Great tit, BT = Blue tit, NH = Nuthatch etc.), and then records the number of eggs, number of chicks, activity of the adults and so on. We need to convert this huge dataset into one which contains clutch size for each nest, for each year of the study.

The information given by `str` (above) shows that there are data on species other than our target species. We are only interested in the great tits and blue tits so we can first filter the others out using the `species` variable.

We can check what the make up of this part of the data is using the `table` function which will count up all of the entries.

```
table(df$species)
```

```
##
##   BT   GT   MT   NH   WR
##  992 4612   73   31    5
```

8.2 filter

Now let's filter this data and double check that this has worked:

```
df <- filter(df, species %in% c("GT", "BT"))
table(df$species)
```

```
##
##   BT   GT
##  992 4612
```

You will notice that all the levels of the variable are retained. This is not a problem, and can usually be ignored. You can also tidy this up using the `droplevels` function, which removes all unused factor levels.

```
df <- droplevels(df)
table(df$species)
```

```
##
##   BT   GT
##  992 4612
```

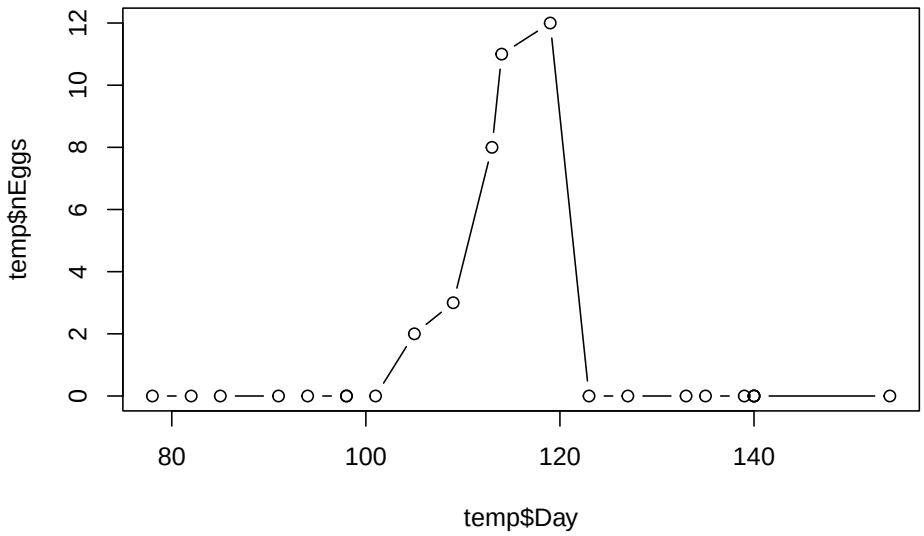
8.3 arrange

Recall that the data are records of visits to each nest a few times per week. To ensure that the data are in time order I can first `arrange` by first `Year` and then `Day`. To illustrate this we can make a temporary data set (called `temp`) to look at a particular nest in a particular year to get a record of the progress for that particular nest, and then plot it (this is an ugly plot and we will learn how to make beautiful ones soon):

```
df <- arrange(df, Year, Day)

temp <- filter(df, boxNumber == 1, Year == 2014)
max(temp$nEggs) # get the max value
```

```
## [1] 12
```

```
plot(temp$Day, temp$nEggs, type = "b")
```



Eggs are usually laid one per day, and the clutch size is the maximum number of eggs reached for each nest box. In this case, the clutch size is 12 eggs. The rapid decline in number of eggs after this peak value shows when the eggs have hatched and the researcher finds chicks instead of eggs!

8.4 summarise and group_by

The next part is the crucial part of our investigation. We need to get the maximum number of eggs seen at each nest. Of course we could repeatedly use `filter`, followed by `max`, for each nest-year combination but this would be incredibly tedious.

Instead, we will use the `dplyr` function, `summarise`, to do this by asking for the maximum value of `nEggs`. To make this work we need to first use the `group_by` function tell R to group the data by the variables we are interested in. If we don't do this we just get the overall maximum. We can ungroup the data using the `ungroup` function.

Because there are missing data (`NA` values) we need to specify `na.rm = TRUE` in the argument.

So first, let's get the max per species, just to illustrate how this works:

```
df <- group_by(df, species)
summarise(df, clutchSize = max(nEggs, na.rm = TRUE))
```

```
## # A tibble: 2 x 2
##   species clutchSize
##   <chr>        <int>
## 1 BT              14
## 2 GT              14
```

We can see how the data are grouped by asking for a summary:

```
summary(df)
```

```
##    species               Year           Day          boxNumber
##  Length:5604        Min.   :2013   Min.   : 60.0   Min.   :  1.00
##  Class :character   1st Qu.:2014   1st Qu.:116.0   1st Qu.: 29.75
##  Mode  :character   Median :2014   Median :133.0   Median : 57.00
##                     Mean   :2015   Mean   :133.2   Mean   : 55.83
##                     3rd Qu.:2017   3rd Qu.:148.0   3rd Qu.: 85.00
##                     Max.   :2019   Max.   :212.0   Max.   :101.00
##
##      nEggs
##  Min.   : 0.000
##  1st Qu.: 0.000
##  Median : 0.000
##  Mean   : 2.326
##  3rd Qu.: 4.000
##  Max.   :14.000
##  NA's   :613
```

We can ungroup the data again like this:

```
df <- ungroup(df)
```

So both species lay the same maximum number of eggs, but maybe this is just caused by outliers for one of the species. We'll need to dig deeper.

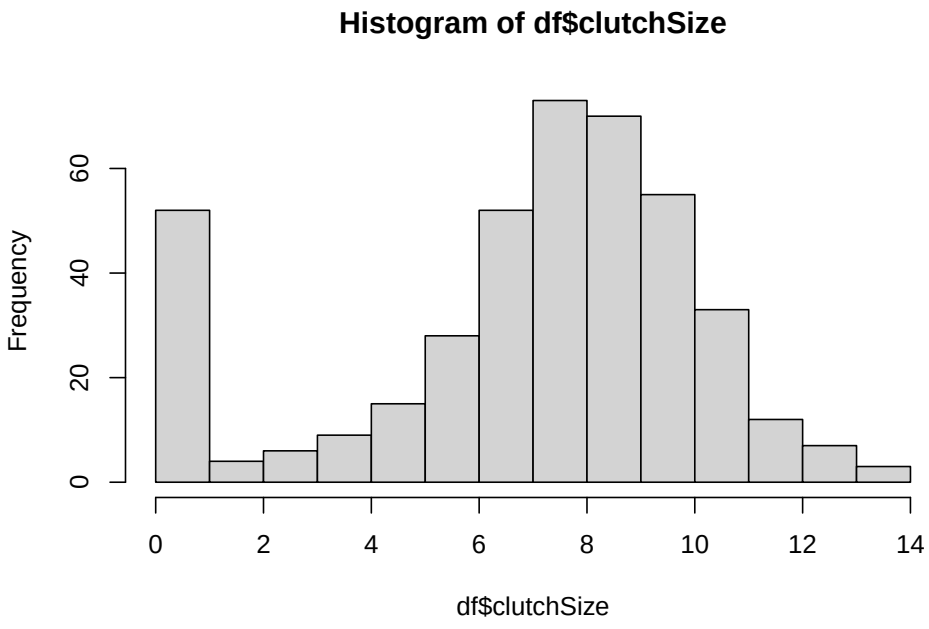
How can we calculate the average? We cannot simply ask for the `mean` because the data run through time following the development in each nest. We need to calculate the maximum `nEggs` for each nest, and then calculate the average of those. We can do this in two steps.

We first calculate the clutch size for each box for each species in each year:

```
df <- group_by(df, species, Year, boxNumber)
df <- summarise(df, clutchSize = max(nEggs, na.rm = TRUE))
```

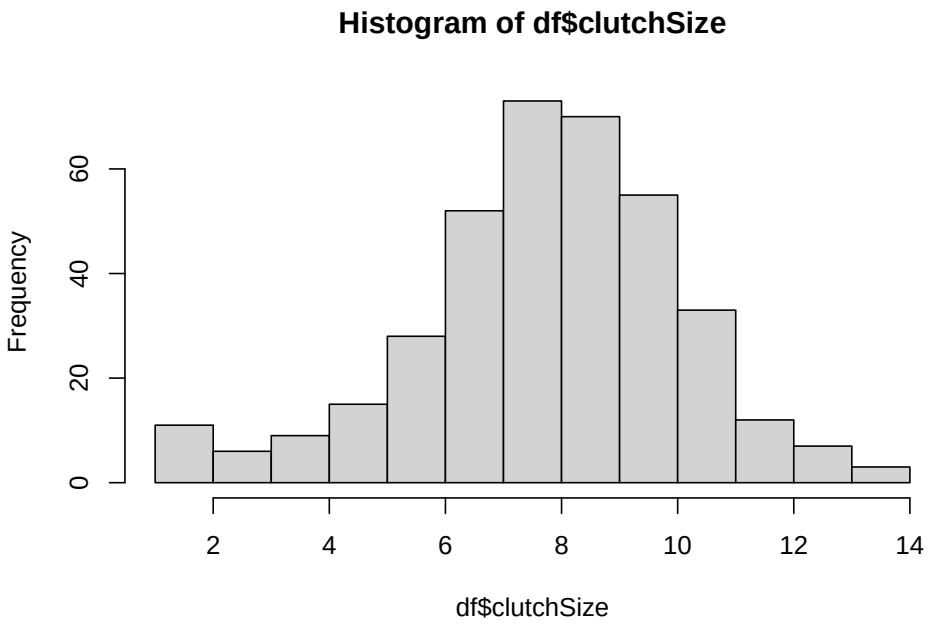
Let's first look at all the clutch size data:

```
hist(df$clutchSize)
```



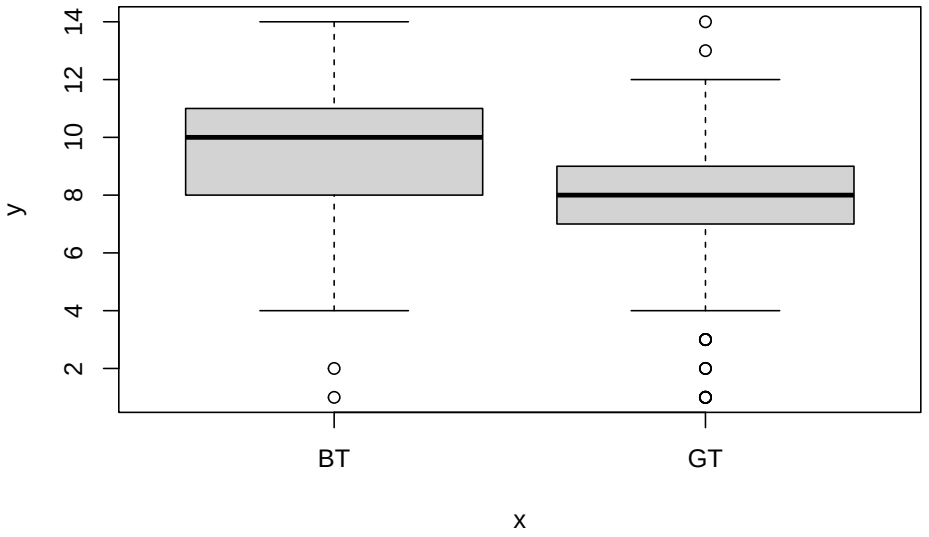
You can see here that there are a lot of zero values. This is because nests were recorded even if they did not attempt to lay eggs. We should remove these from our data using `filter` again:

```
df <- filter(df, clutchSize > 0)
hist(df$clutchSize)
```



That looks better. Now we can plot them again but this time split apart the species (again - this plot is ugly and we'll learn to plot nicer ones soon).

```
plot(as.factor(df$species), df$clutchSize)
```



From these distributions it looks like the average clutch size is greater in the blue tit. We can use `summarise` to calculate the means.

```
df <- group_by(df, species)
summarise(df, mean = mean(clutchSize), sd = sd(clutchSize))
```

```
## # A tibble: 2 x 3
##   species  mean    sd
##   <chr>   <dbl> <dbl>
## 1 BT       9.64  2.64
## 2 GT       7.89  2.19
```

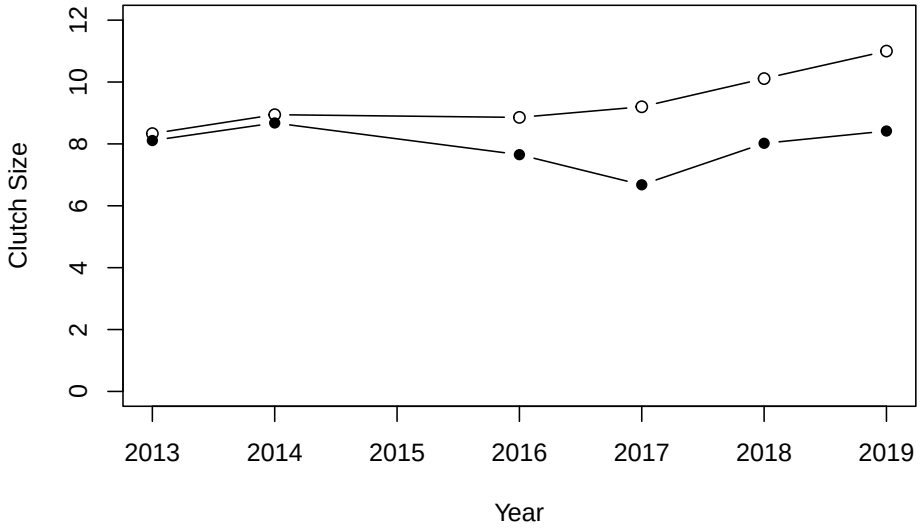
Lets now turn to the other question - how does the clutch size vary with year?

```
df <- group_by(df, species, Year)
df2 <- summarise(df, meanClutchSize = mean(clutchSize))
head(df2)
```

```
## # A tibble: 6 x 3
## # Groups:   species [1]
##   species  Year meanClutchSize
##   <chr>   <int>          <dbl>
## 1 BT       2013           8.33
## 2 BT       2014           8.94
## 3 BT       2016           8.86
## 4 BT       2017           9.2
## 5 BT       2018          10.1
## 6 BT       2019          11
```

We can plot this by first making a plot for blue tits, and then adding the points for great tits. I have used the `pch` argument to use filled circles for the great tits:

```
plot(df2$Year[df2$species == "BT"],
  df2$meanClutchSize[df2$species == "BT"],
  type = "b",
  ylim = c(0, 12), xlab = "Year", ylab = "Clutch Size"
)
points(df2$Year[df2$species == "GT"],
  df2$meanClutchSize[df2$species == "GT"],
  pch = 16, type = "b"
)
```



So it looks like the clutch size varies a fair amount from year to year, but that generally blue tits have large clutch sizes than great tits.

8.5 Using pipes, saving data.

I have walked you through a step-by-step data manipulation. During that process you made made (and replaced) new data sets at each step. In practice this can be done more smoothly using *pipes* (%>%) to pass the result of one function into the next, and the next, and the next…

You'll get some more practice with this as we go on. Below I show how do do this to create a clutch size data set from the raw data (note that your file path will differ from mine):

```
# Import and process data using pipes
SDUClutchSize <- read.csv("CourseData/sduBirds.csv") %>%
  filter(species %in% c("GT", "BT")) %>% # include only GT and BT
  droplevels() %>% # drop unwanted factor levels
  select(species, Year, Day, boxNumber, nEggs) %>% # select columns
  group_by(species, Year, boxNumber) %>% # group data
  summarise(clutchSize = max(nEggs, na.rm = TRUE)) %>% # clutch size
  filter(clutchSize > 0)

head(SDUClutchSize)
```

```
## # A tibble: 6 x 4
## # Groups:   species, Year [1]
##   species  Year boxNumber clutchSize
##   <chr>   <int>     <int>      <dbl>
## 1 BT       2013         1         12
```

```
## 2 BT       2013          5          8
## 3 BT       2013         27         10
## 4 BT       2013         31          6
## 5 BT       2013         35         10
## 6 BT       2013         37          8
```

You can save this out using the `write.csv` function. You will need to set the argument `row.names = FALSE` to stop the data including row names.

```
write.csv(x = SDUClutchSize, file = "CourseData/SDUClutchSize.csv",
          row.names = FALSE)
```

8.6 Exercise: Wrangling the Amniote Life History Database

In this exercise the aim is to use the "Amniote Life History Database" [1] to investigate some questions about life history evolution.

The questions are: (1) what are the records and typical life spans in different taxonomic classes? [*what is the longest, shortest and median life span in birds, mammals and reptiles?*] (2) is there a positive relationship between body mass and life span? [*do big species live longer than small ones?*]; (3) is there a trade-off between reproductive effort and life span? [*do species that reproduce a lot have short lives, so there is a negative relationship between reproduction and life span?*]; (4) is this trade-off universal across all Classes? [*does the trade-off exist in birds, reptiles and amphibians?*]

The database is in a file called `Amniote_Database_Aug_2015.csv` in the course data folder. The missing values (which are normally coded as `NA` in R) are coded as "`-999`". The easiest way to take care of this is to specify this when we import the data using the `na.strings` argument of the `read.csv` function. Thus we can import the data like this:

```
amniote <- read.csv("CourseData/Amniote_Database_Aug_2015.csv",
                    na.strings = "-999")
```

Let's make a start…

1. When you have imported the data, use `dim` to check the dimensions of the whole data frame (you should see that there are 36 columns and 21322 rows). Use `names` to look at the names of all columns in the data in `amniote`.

[1] https://esajournals.onlinelibrary.wiley.com/doi/10.1890/15-0846R.1

2. We are interested in longevity (lifespan) and body size and reproductive effort and how this might vary depending on the taxonomy (specifically, with Class). Use `select` to pick relevant columns of the dataset and discard the others. Call the new data frame `x`. The relevant columns are the taxonomic variables (`class`, `genus` & `species`) and `longevity_y`, `litter_or_clutch_size_n`, `litters_or_clutches_per_y`, and `adult_body_mass_g`.

3. Take a look at the first few entries in the `species` column. You will see that it is only the *epithet*, the second part of the *Genus_species* name, that is given.
   Use `mutate` and `paste` to convert the `species` column to a *Genus_species* by pasting the data in `genus` and `species` together. To see how this works, try out the following command, `paste(1:3, 4:6)`. After you have created the new column, remove the `genus` column (using `select` and `-genus`).

4. What is the longest living species in the record? Use `arrange` to sort the data from longest to shortest longevity (`longevity_y`), and then look at the top of the file using `head` to find out. (hint: you will need to use reverse sort (`-`)). Cut and paste the species name into Google to find out more!

5. Do the same thing but this time find the shortest lived species.

6. Use `summarise` and `group_by` to make a table summarising `min`, `median` and `max` life spans (`longevity_y`) for the three taxonomic classes in the database. Remember that you need to tell R to remove the `NA` values using a `rm.rm = TRUE` argument.

7. Body size is thought to be associated with life span. Let's treat that as a hypothesis and test it graphically. Sketch what would the graph would look like if the hypothesis were true, and if it was false. Plot `adult_body_mass_g` vs. `longevity_y` (using base R graphics). You should notice that this looks a bit messy.

8. Use `mutate` to create new `log`-transformed variables, `logMass` and `logLongevity`. Use these to make a "log-log" plot. You should see that makes the relationship more linear, and easier to "read".

9. Is there a trade-off between reproductive effort and life span? Think about this as a hypothesis - sketch what would the graph would look like if that were true, and if it was false. Now use the data to test that hypothesis: Use `mutate` to create a variable called `logOffspring` which is the logarithm of number of litters/clutches per year multiplied by the number of babies in each litter/clutch . Then plot `logOffspring` vs. `logLongevity`.

10. To answer the final question (differences between taxonomic classes) you could now use `filter` to subset to particular classes and repeat the plot to see whether the relationships holds universally.

Remember that if you struggle you can check back to previous work where you have used `dplyr` commands to manipulate data in a similar way. If you get truly stuck, ask for help from instructors or fellow students.

Chapter 9

Combining data sets

Handling data is often not limited to single data sets. One common task is to combine two (or more) datasets together. For example, one dataset might include a set of observations from a field study, while another might have information about the weather throughout the study period, or site-specific information. It is therefore useful to be able to combine these datasets to add the information from the second table to the first table.

R does this with the `dplyr` function `join`. In the next section you will first learn how `join` works by following an example that asks a research question that can only be answered by two data sets. After that, you will work on your own to do a similar analysis without explicit instructions (i.e. you will need to figure out how to apply the method to new data.

9.1 Using `join`

By following this example you will learn how to combine two data sets to create a new one of combined data to answer a conservation-related question: "**Does threat status vary with species' generation times?**"

This question is crucial to conservation biologists because it helps us to generalise our ideas about what drives extinction risks. In other words, if we can say "*species with slow life histories tend to be more threatened*" then this gives useful information that can help with planning. For example, imagine we have some species that have not yet been assessed (we don't know if they are threatened or not). Should we focus attention on the one with a short generation time, or long generation time?

To answer the question we will need to import two large data sets, tidy them up a bit and then combine them for analysis.

Let's start with the "Amniote Life History Database" [1], which is a good source of life history data. We have encountered this database before. Recall that the

[1] https://esajournals.onlinelibrary.wiley.com/doi/10.1890/15-0846R.1

missing values (which are normally coded as NA in R) are coded as "-999". The easiest way to take care of this is to specify this when we import the data using the na.strings argument of the read.csv function. Thus we can import the data like this:

```
amniote <- read.csv("CourseData/Amniote_Database_Aug_2015.csv",
  na.strings = "-999"
)
```

We can filter on the taxonomic class to subset to only mammals. Then, to address our question, we want data on generation time for mammals. Generation time is often measured as the average age at which females reproduce so we can get close to that with female_maturity_d. We will first select these columns, along with genus and species. We can combine these two taxonomic variables using mutate and paste to get our Latin binomial species name.

We have previously learned that log transforming such variables is a good thing to do, so we can use mutate again to do this transformation.

Finally, we can use na.omit to get rid of entries with missing values (which we cannot use). This is not essential, but keeps things more manageable.

```
mammal <- amniote %>%
  filter(class == "Mammalia") %>%
  # get the mammals only
  select(genus, species, female_maturity_d) %>%
  # get useful columns
  mutate(species = paste(genus, species)) %>%
  select(-genus) %>%
  mutate(logMaturity = log(female_maturity_d)) %>%
  na.omit()
```

Let's take a quick look at what we have:

```
head(mammal)
```

```
##                       species female_maturity_d logMaturity
## 20          Echinops telfairi         278.42000    5.629131
## 22    Hemicentetes nigriceps          48.57000    3.883006
## 23 Hemicentetes semispinosus          46.19892    3.832956
## 27          Microgale dobsoni         669.59200    6.506669
## 40         Microgale talazaci         639.00000    6.459904
## 47            Setifer setosus         198.00000    5.288267
```

Looks good. Now let's import the IUCN Red List data.

```
redlist <- read.csv("CourseData/MammalRedList.csv")
```

Let's take a look at that.

```
names(redlist)
```

```
##  [1] "Species.ID"                "Kingdom"
##  [3] "Phylum"                    "Class"
##  [5] "Order"                     "Family"
##  [7] "Genus"                     "Species"
##  [9] "Authority"                 "Infraspecific.rank"
## [11] "Infraspecific.name"        "Infraspecific.authority"
## [13] "Stock.subpopulation"       "Synonyms"
## [15] "Common.names..Eng."        "Common.names..Fre."
## [17] "Common.names..Spa."        "Red.List.status"
## [19] "Red.List.criteria"         "Red.List.criteria.version"
## [21] "Year.assessed"             "Population.trend"
## [23] "Petitioned"
```

```
unique(redlist$Red.List.status)
```

```
## [1] "DD" "LC" "CR" "NT" "EN" "VU" "EX" "EW"
```

There's a lot of information there but what we really need is simply the Latin binomial (for which we need `genus` and `species`) and the threat status `Red.List.status`.

R treats categorical variables (`factor` variables) as alphabetical, but in this case the red list status has a meaning going from low threat (Least Concern - LC) to Critically Endangered (CR) and even Extinct in the Wild (EX) at the other end of the spectrum. We can define this ordering using `mutate` with the `factor` function.

```
redlist <- redlist %>%
  mutate(species = paste(Genus, Species)) %>%
  select(species, Red.List.status) %>%
  mutate(Red.List.status = factor(Red.List.status,
    levels = c("LC", "NT", "VU", "EN", "CR", "EW", "EX")
  ))

head(redlist)
```

```
##                species Red.List.status
## 1   Abditomys latidens            <NA>
```

```
## 2    Abeomelomys sevia              LC
## 3 Abrawayaomys ruschii              LC
## 4   Abrocoma bennettii              LC
## 5 Abrocoma boliviensis              CR
## 6      Abrocoma budini            <NA>
```

Now we can combine this with the life history data from above using `left_join`.

```
x <- left_join(mammal, redlist, by = "species")
```

Let's take a look at what we have now:

```
head(x)
```

```
##                     species female_maturity_d logMaturity Red.List.status
## 1         Echinops telfairi         278.42000    5.629131              LC
## 2    Hemicentetes nigriceps          48.57000    3.883006              LC
## 3 Hemicentetes semispinosus          46.19892    3.832956              LC
## 4         Microgale dobsoni         669.59200    6.506669              LC
## 5        Microgale talazaci         639.00000    6.459904              LC
## 6           Setifer setosus         198.00000    5.288267              LC
```

```
summary(x)
```

```
##    species          female_maturity_d  logMaturity    Red.List.status
##  Length:2000        Min.   :  23.81   Min.   :3.170   LC     :1219
##  Class :character   1st Qu.: 121.53   1st Qu.:4.800   VU     : 176
##  Mode  :character   Median : 344.12   Median :5.841   EN     : 168
##                     Mean   : 574.92   Mean   :5.745   NT     : 114
##                     3rd Qu.: 696.38   3rd Qu.:6.546   CR     :  66
##                     Max.   :6391.56   Max.   :8.763   (Other):  10
##                                                       NA's   : 247
```

You can see that there are 247 missing values for the Red List status. These are either species that have not yet been assessed, or maybe where there are mismatches in the species names between the two databases. We will ignore this problem today.

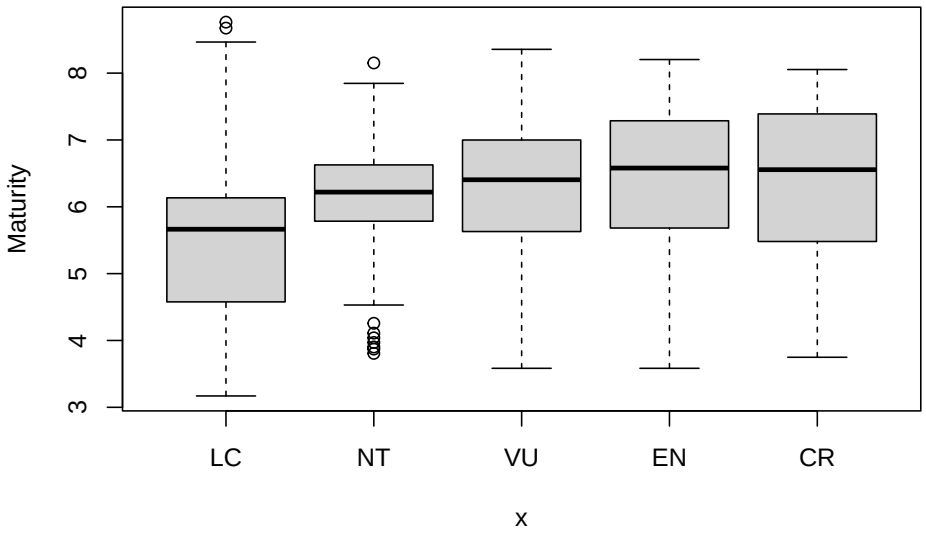
Before plotting, I will also use `filter` remove species that are extinct (status = "EX" and "EW"). To do this I use the `%in%` argument to allow me to match a vector of variables. Because I want to NOT match them I negate the match using `!`.

I then ensure that those levels are removed from the variable using `droplevels`.

```
x <- x %>%
  filter(!Red.List.status %in% c("EX", "EW")) %>%
  droplevels()
```

Let's now plot the data to answer the question.

```
plot(x$Red.List.status, x$logMaturity, ylab = "Maturity")
```



What can we see? If you focus on the median values, it looks like there is a weak positive relationship between this life history trait and threat status: animals with slower life histories tend to be more threatened.

9.2 Using `pivot_longer`

Sometimes data are not arranged in a way that make them easy to use in R. For example, data could be arranged so that the column headings are themselves data (e.g. treatments, sex of individuals).

To illustrate this I will use a data set on heights of men and women.

```
heights <- read.csv("CourseData/heights.csv")
```

Let's take a look:

```
heights
```

```
##      place heightMale heightFemale
## 1   London        170          164
## 2   London        176          158
## 3   London        179          157
## 4   London        166          158
## 5   London        177          153
## 6   London        177          155
## 7   London        173          151
## 8   London        173          155
## 9   London        173          159
## 10  London        171          158
## 11  London        173          166
## 12  London        171          156
## 13  London        172          157
## 14  London        175          159
## 15  London        179          156
## 16 Bristol        175          156
## 17 Bristol        173          156
## 18 Bristol        171          155
## 19 Bristol        172          158
## 20 Bristol        185          158
## 21 Bristol        176          153
## 22 Bristol        173          158
## 23 Bristol        173          156
## 24 Bristol        177          156
## 25 Bristol        172          159
## 26 Bristol        169          162
## 27 Bristol        177          167
## 28 Bristol        171          157
## 29 Bristol        175          166
## 30 Bristol        171          155
```

I would like to make a boxplot, but it is not possible to easily do it with the data arranged in this format. What I need to do is "unpack" or rearrange the data to add another column for sex. This will make the data frame twice as long, and less wide.

There is a convenient function called `pivot_longer` in the `tidyr` package that will do this for you. You can "pipe" data into the function, then tell it which columns you would like to move, and then give it the name of the new column that contains data that **was** in the column heading, and the name of the column containing the data.

```
newHeights <- heights %>%
  pivot_longer(
    cols = c(heightMale, heightFemale),
    names_to = c("Sex"), values_to = "Height"
  )
```

```
newHeights
```

```
## # A tibble: 60 x 3
##    place  Sex          Height
##    <chr>  <chr>         <int>
##  1 London heightMale      170
##  2 London heightFemale    164
##  3 London heightMale      176
##  4 London heightFemale    158
##  5 London heightMale      179
##  6 London heightFemale    157
##  7 London heightMale      166
##  8 London heightFemale    158
##  9 London heightMale      177
## 10 London heightFemale    153
## # ... with 50 more rows
```

We are nearly done. But this is not perfect because the names in the `Sex` column are not right. We can fix this in a couple ways. Here's one easy way using the function `gsub`. The `gsub` function ("**g**eneral **sub**stution) finds text and replaces it with other text. In this case we want to find"height" and replace it with nothing ("").

So we can now complete the job with a `mutate` command, and make sure it is recognised as a categorical variable (a `factor`, like this:

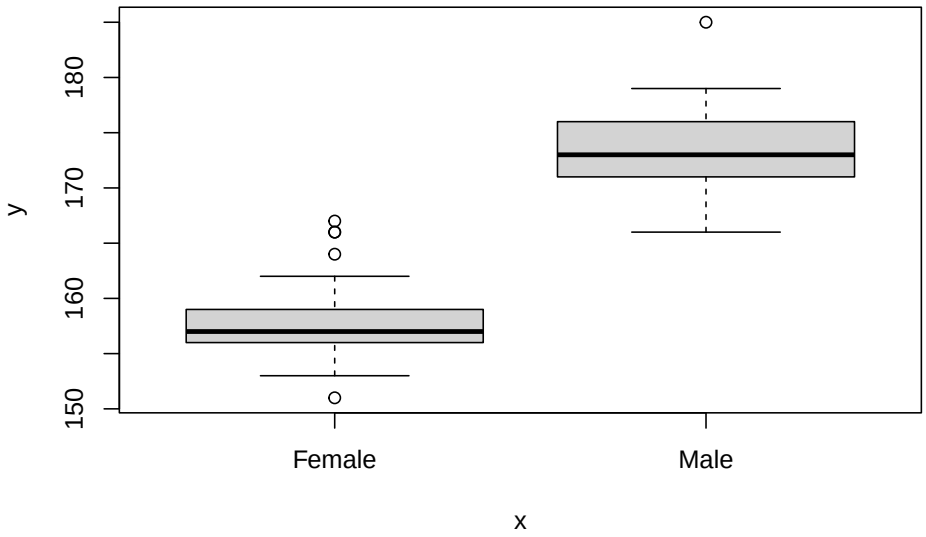
```
newHeights <- heights %>%
  pivot_longer(
    cols = c(heightMale, heightFemale),
    names_to = c("Sex"), values_to = "Height"
  ) %>%
  mutate(Sex = gsub(
    pattern = "height", replacement = "",
    x = Sex
  )) %>%
  mutate(Sex = as.factor(Sex))

newHeights
```

```
## # A tibble: 60 x 3
##    place  Sex    Height
##    <chr>  <fct>   <int>
##  1 London Male      170
##  2 London Female    164
##  3 London Male      176
##  4 London Female    158
##  5 London Male      179
```

```
##  6 London Female    157
##  7 London Male      166
##  8 London Female    158
##  9 London Male      177
## 10 London Female    153
## # ... with 50 more rows
```

Now we can plot those data more easily

```
plot(newHeights$Sex, newHeights$Height)
```



This data manipulation is useful surprisingly often.

9.3 Exercise: Temperature effects on egg laying dates

Data have been collected on great tits (musvit) at SDU for several years. Your task today is to analyse these data to answer the question: *is egg laying date associated with spring temperature?* The idea here is that warmer springs will lead to delayed egg laying which could have negative consequences to the population if their caterpillar food source doesn't keep pace with the change.

You are provided with two data sets: one on the birds and another on weather. You will need to process these using tools in the `dplyr` package, and combine them (using `left_join`) for analysis.

The first data set, `eggDates.csv`, is data from the SDU birds project. The data are arranged in columns where each column is a year and each row is a nest. The data in each column is the day of the year that the first egg in the nest was laid.

These data do NOT fulfil the "tidy data" standard where each variable gets a column. In this case,a single variable (first egg date) gets many columns (one for each year), and column headers are data (the years). The data will need to be processed before you can analyse it.

You will need to use `pivot_longer` to fix this issue so that you produce a version of the data with three columns - `nestNumber`, `Year` and `dayNumber`.

The second dataset, `AarslevTemperature.csv`, is a weather dataset from Årslev near Odense. This dataset includes daily temperatures records for several years. You will need to `summarise` this data to obtain a small dataset that has the weather of interest - average temperature in the months of February to April for each year.

To answer the question, you will need to join these data sets together.

1. Import the data and take a look at it with `head` or `str`.
2. Use `pivot_longer` to reformat the data. This might take a bit of trial and error - don't give up!

Maybe this will help: The first argument in the `pivot_longer` command (`cols`) tells R which columns contain the data you are interested in (in this case, these are `y2013`,`y2014` etc). Then the `names_to` argument tells R what you want to name the new column from this data (in this case, `Year`). Then, the `values_to` argument tells R what the data column should be called (e.g. `Day`). In addition, there is a useful argument called `names_prefix` that will remove the part of the column name (e.g. the `y` of `y2013`)

You should also make sure that the `Year` column is recognised as being a numeric variable rather than a character string. You can do this by adding a command using `mutate` and `as.numeric`, like this `mutate(Year = as.numeric(Year))`

You should end up with a dataset with three columns as described above.

3. Calculate the mean egg date per year using `summarise` (remember to `group_by` the year first). Take a look at the data.
4. Import the weather data and take a look at it with `head` or `str`.
5. Use `filter` subset to the months of interest (February-April) and then `summarise` the data to calculate the mean temperature in this period (remember to `group_by` year). Look at the data. You should end up with a dataset with two columns - `year` and `meanSpringTemp`.
6. Join the two datasets together using `left_join`. You should now have a dataset with columns `nestNumber`, `Year`, `dayNumber` and `meanAprilTemp`
7. plot a graph of `meanAprilTemp` on the x-axis and `dayNumber` on the y-axis.

Now you should be able to answer the question we started with: is laying date associated with spring temperatures.

Part II

Data visualisation

Chapter 10

Visualising data with ggplot

In this chapter you will be guided through using the `ggplot2` package to make some pretty plots. You will therefore need the `ggplot2` package to make this work. Remember, you can load packages like this:

```
library(ggplot2)
```

We will use the SDU birds clutch size data that we produced at the end of the "[Data wrangling with dplyr]" chapter for these examples. You can find the data set via the Course Data Dropbox link.

> Remember to set your working directory, and start a new script. I am assuming that you have saved your data in a folder called "CourseData" inside your working directory.

```
df <- read.csv("CourseData/SDUClutchSize.csv")
```

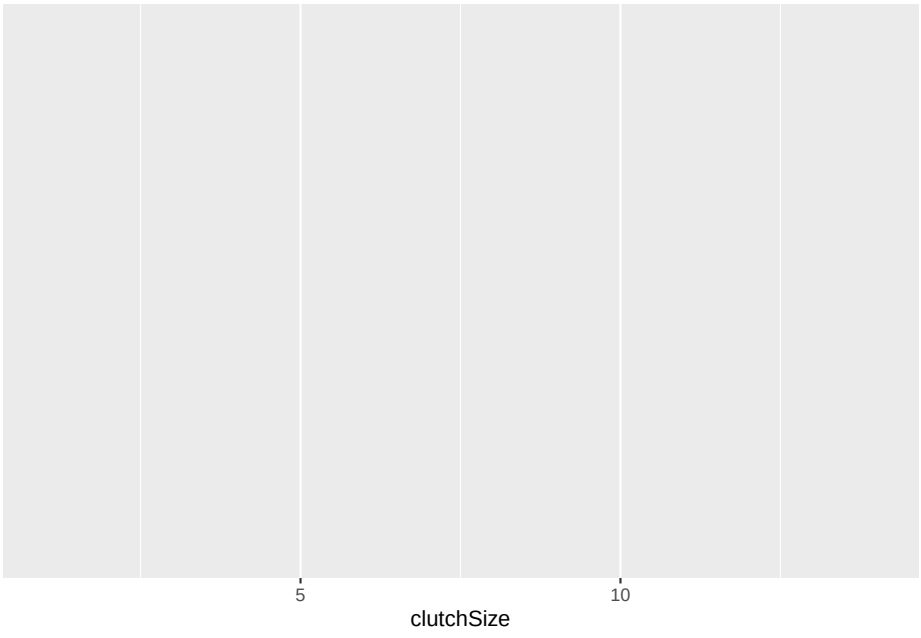
10.1 Histograms

The `ggplot` function expects two main arguments (1) the data and (2) the **aesthetics**. The aesthetics are the variables you want to plot, and associated characteristics like colours, groupings etc. The first argument is for the data, then the aesthetics are specified within the `aes(...)` argument. These usually include an argument for `x` which is normally the variable that appears on the horizontal axis, and (often) `y` which is usually the variable on the vertical axis. The details of this depend on the type of plot you are making.

After setting up the plot the graphics are added as **geometric layers** or **geoms**. There are many of these available including `geom_histogram`, `geom_line`, `geom_point` etc.

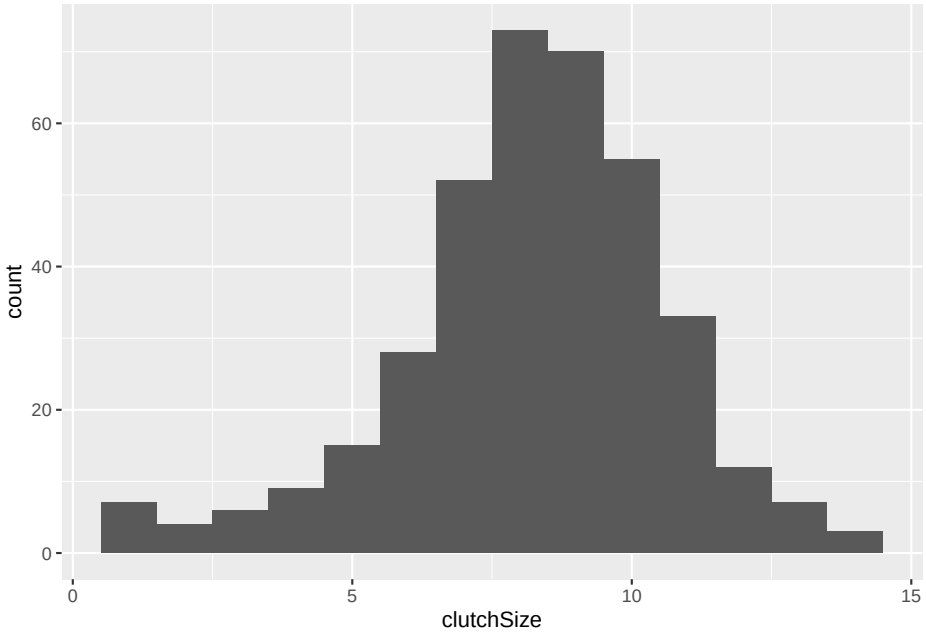
I will illustrate the construction of a simple plot by making a histogram of the clutch size of all the nests in the dataset.

```
ggplot(df, aes(x = clutchSize))
```



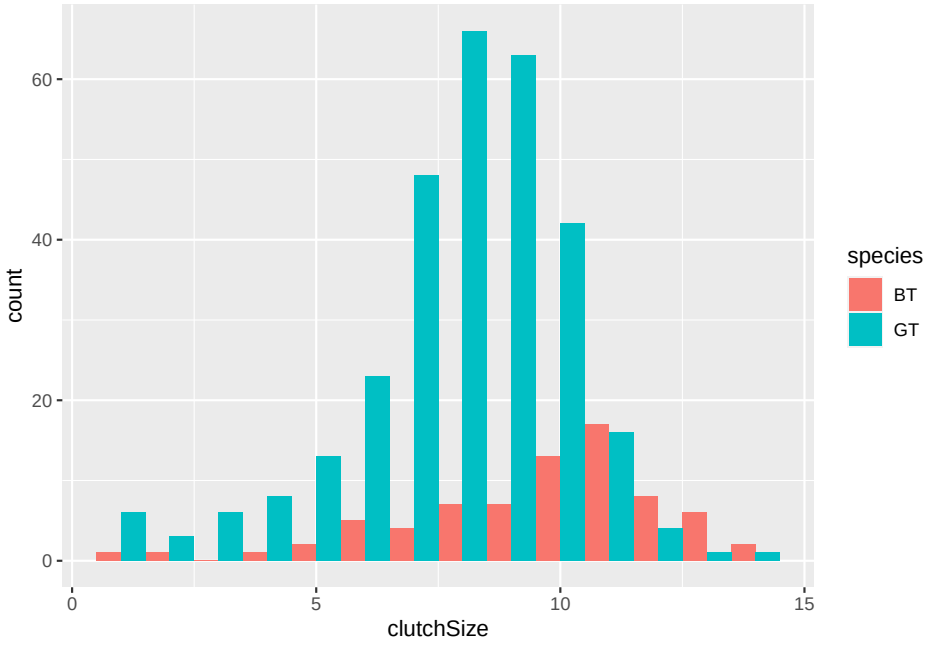
This produces an empty plot because we have not yet specified what kind of plot we want. We want a histogram, so we can add this as follows. I have set `binwidth` to be 1 because we know we are dealing with counts between just 1 and 14. Try altering the `binwidth`.

```
ggplot(df, aes(x = clutchSize)) +
  geom_histogram(binwidth = 1)
```



We know that we have two species here and we would like to compare them. This is done within the aesthetic argument. The default is that the bars for different categories are stacked on top of each other. This is good in some cases, but probably not here.

```
ggplot(df, aes(x = clutchSize, fill = species)) +
  geom_histogram(binwidth = 1, position = "dodge")
```

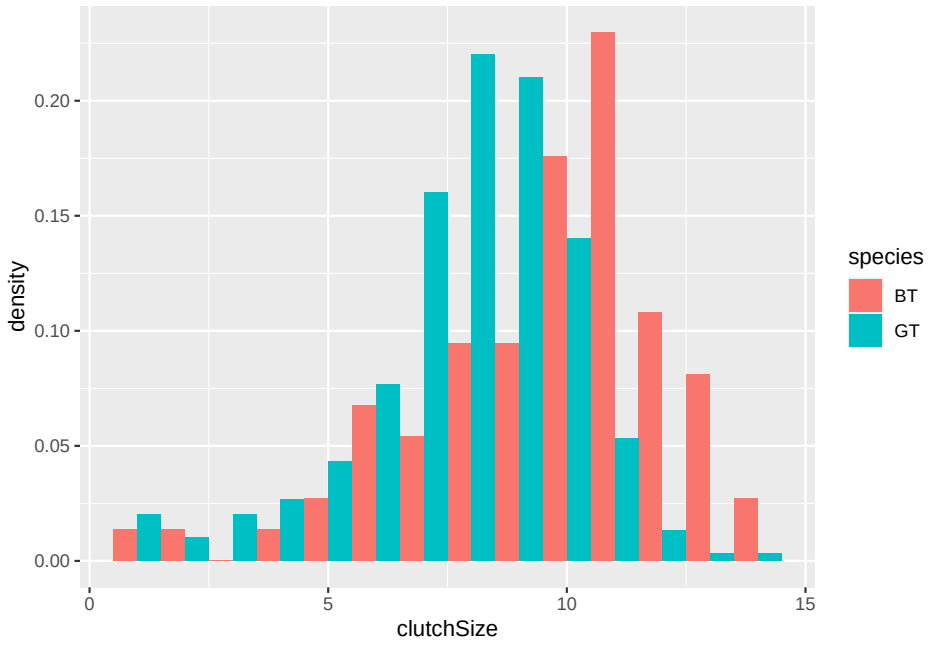


You can immediately see that there are far fewer blue tit nests than great tit

ones. But you can also see that the centre of mass for blue tits is further to the right than great tits.

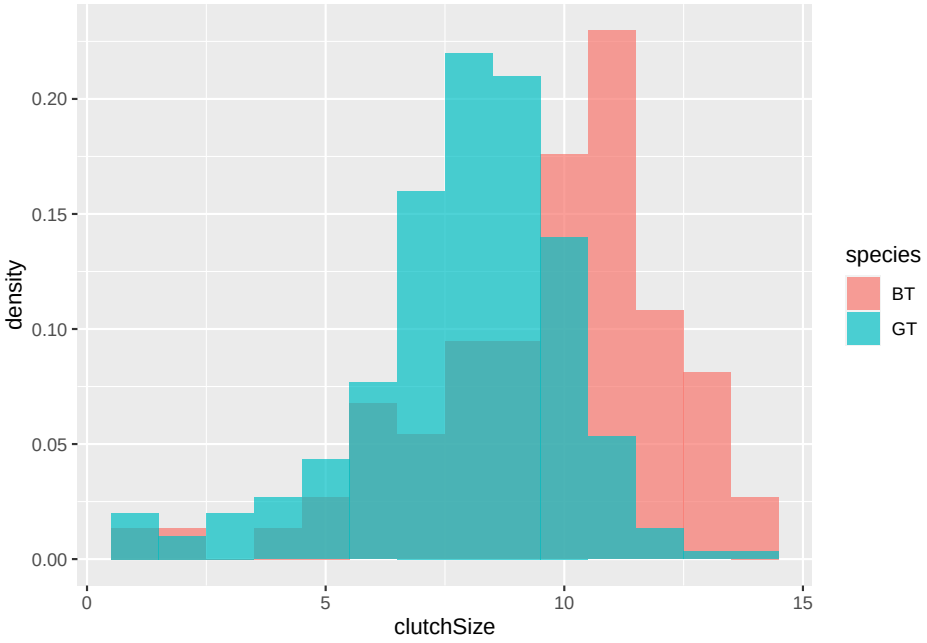
To make it easier to compare distributions with very different counts, we can put density on the y-axis instead of the default count using the argument `stat(density)`.

```
ggplot(df, aes(x = clutchSize, fill = species, stat(density))) +
  geom_histogram(binwidth = 1, position = "dodge")
```



An alternative approach would be to overlay the two sets of bars (using `position = "identity"`) and set the colours to be slightly transparent (using `alpha = 0.7`) so that you can see the overlapping region clearly.

```
ggplot(df, aes(x = clutchSize, fill = species, stat(density))) +
  geom_histogram(binwidth = 1, position = "identity", alpha = 0.7)
```

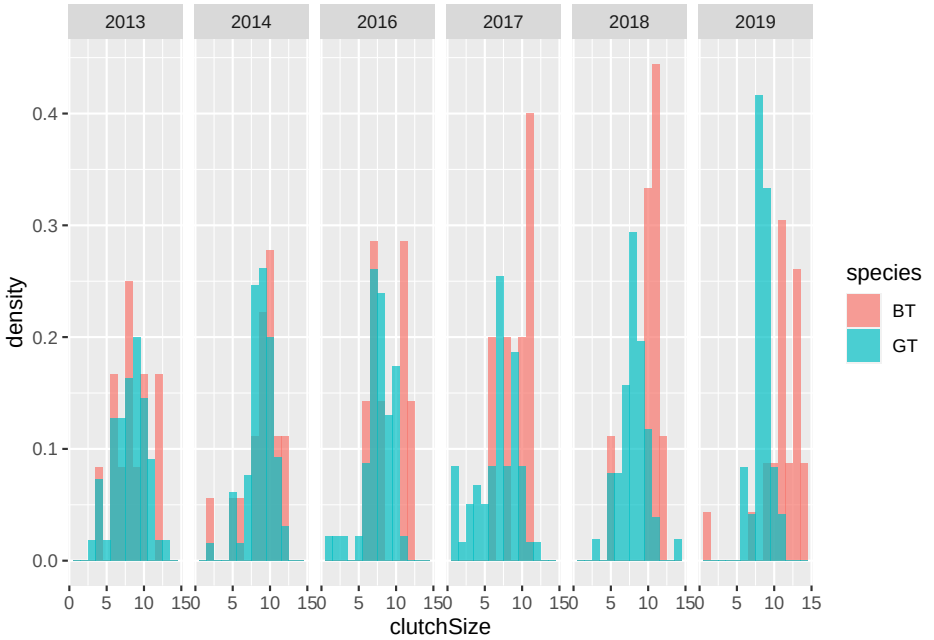


It is very clear from this plot that blue tits tend to have bigger clutch sizes than great tits. Is this difference *statistically significant*? We will look at testing this in a future class - for now we will be satisfied with our visualisation.

10.2 "Facets" - splitting data across panels

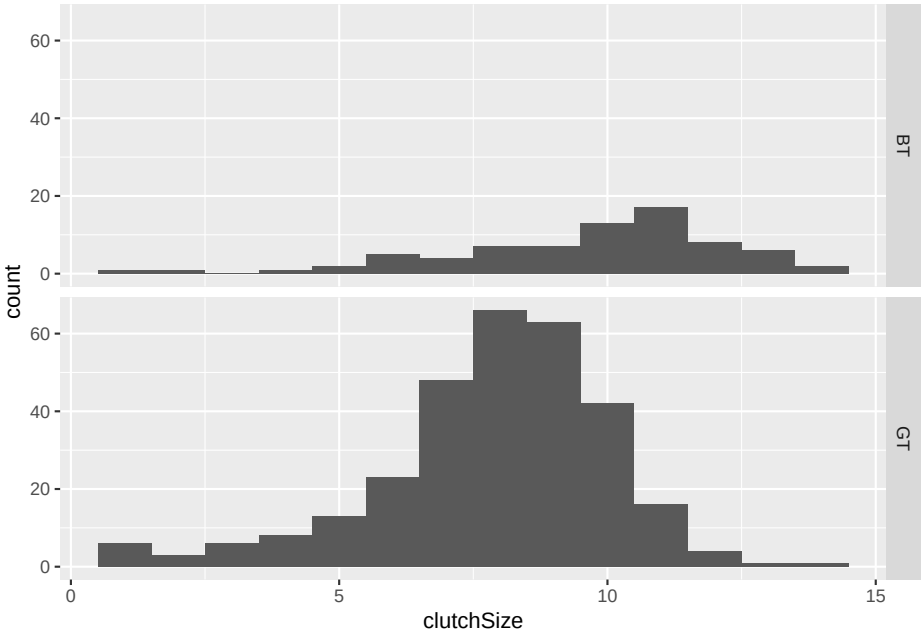
You should recall that there were several years of data represented here. `ggplot` has a very clever way of splitting up the plot to examine this.

```
ggplot(df, aes(x = clutchSize, fill = species, stat(density))) +
  geom_histogram(
    binwidth = 1, position = "identity",
    alpha = 0.7
  ) +
  facet_grid(. ~ Year)
```



You could split the data up by species in a similar way, as yet another way of visualising the difference between species:

```
ggplot(df, aes(x = clutchSize)) +
  geom_histogram(binwidth = 1) +
  facet_grid(species ~ .)
```



You can change whether the separate graphs are presented in a rows or columns by changing the order of the argument: `facet_grid(species~.)` or `facet_grid(.~species)`. Try it.

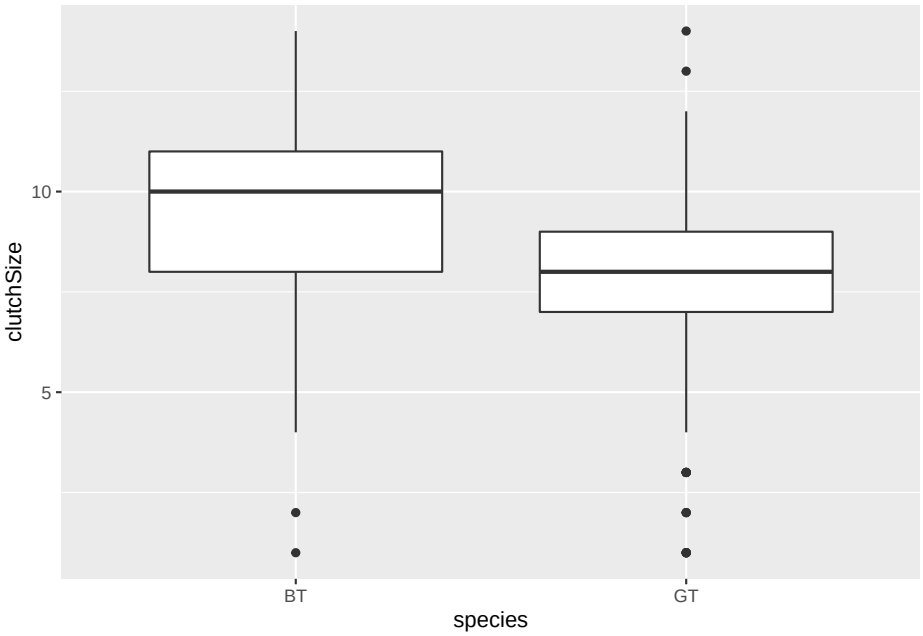
10.3 Box plots

Box plots are suitable for cases where one variable is categorical with 2+ levels, and the other is continuous. Therefore, another way to look at these distributions is to use a box plot.

In a box plot the box shows the quartiles (i.e. the 25% and 75% quantiles) within which 50% of the data are found. The horizontal line in the box is the *median*, Then the whiskers extend from the smallest to largest value *unless they are further than 1.5 times the interquartile range (the length of the box)* away from the edge of the box, in which case they are individually shown as outlier points.

To plot them using `ggplot` you must use a `geom_boxplot` layer. The categorical variable is normally placed on the x-axis so is placed as `x` in the `aes` argument, while the continuous variable is on the `y` axis.

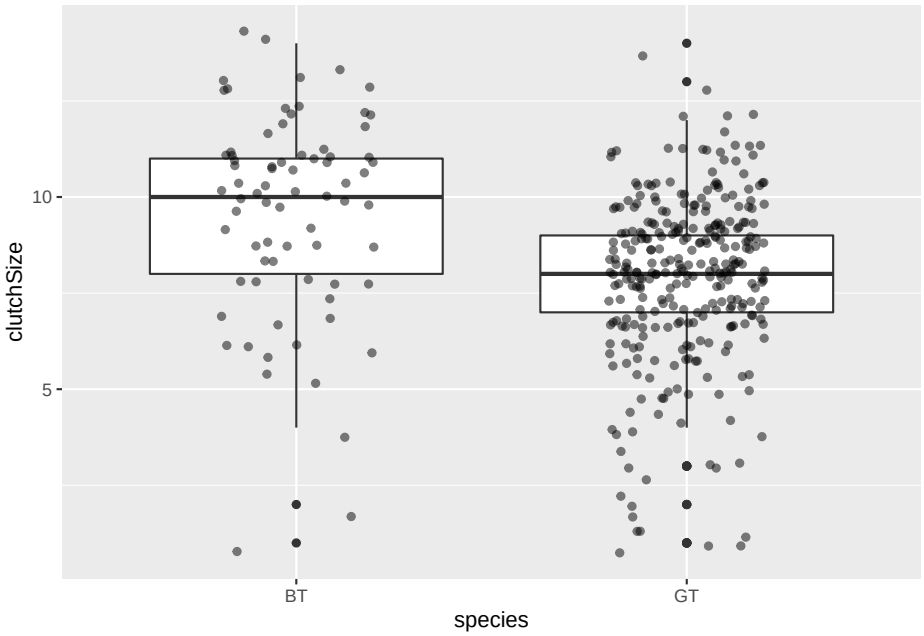
```
ggplot(df, aes(x = species, y = clutchSize)) +
  geom_boxplot()
```



Some researchers argue that it is a good idea to add the data as points to these plots as "full disclosure" of what the underlying data look like. These can be

added with a `geom_jitter` layer (jitter is random noise added in this case to the horizontal axis). You should set `width` and `alpha` arguments to make it look nice.

```
ggplot(df, aes(x = species, y = clutchSize)) +
  geom_boxplot() +
  geom_jitter(width = .2, alpha = 0.5, colour = "black", fill = "black")
```



> Try splitting the data into different years using `facet_grid` with the box plot.

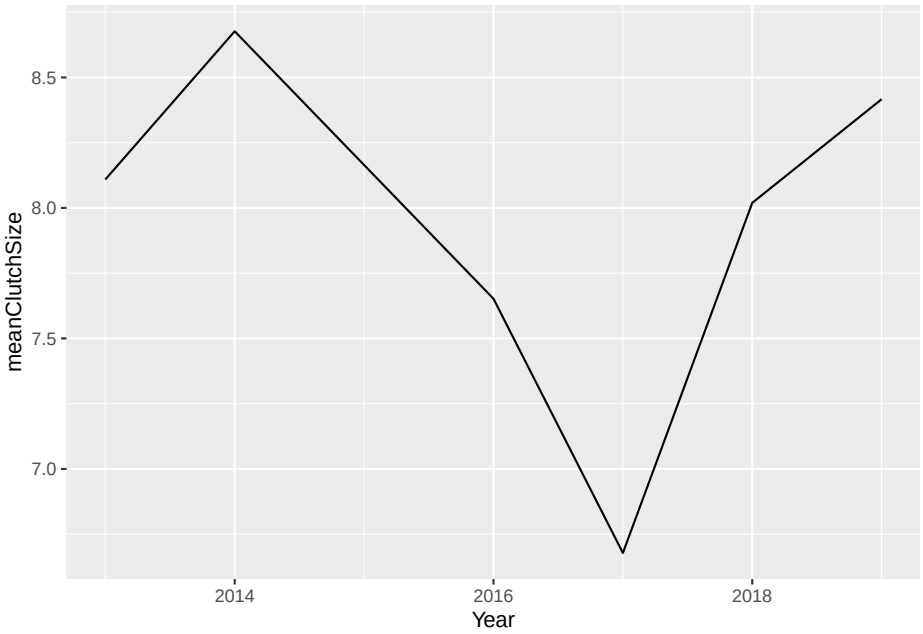
10.4 Lines and points

Perhaps not surprisingly lines and points can be added with the geoms, `geom_line` and `geom_point` respectively. To illustrate this we will make a plot showing how clutch size changes among years. First we will use `summarise` to create a dataset with the mean clutch size. We'll start simply, by looking at only great tits.

```
GTclutch <- df %>%
  filter(species == "GT") %>%
  group_by(Year) %>%
  summarise(meanClutchSize = mean(clutchSize))
```

Then you can plot this like this.

```
ggplot(GTclutch, aes(x = Year, y = meanClutchSize)) +
  geom_line()
```

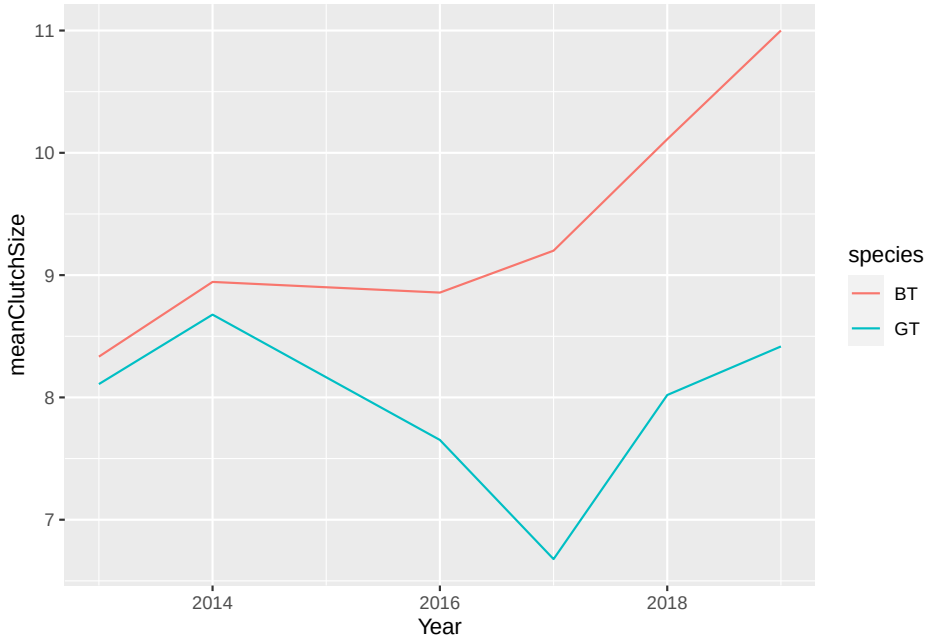


I think this looks OK, but we should add both species. I'll first need to produce a mean clutch size dataset that includes both species.

```
meanClutch <- df %>%
  group_by(species, Year) %>%
  summarise(meanClutchSize = mean(clutchSize))
```

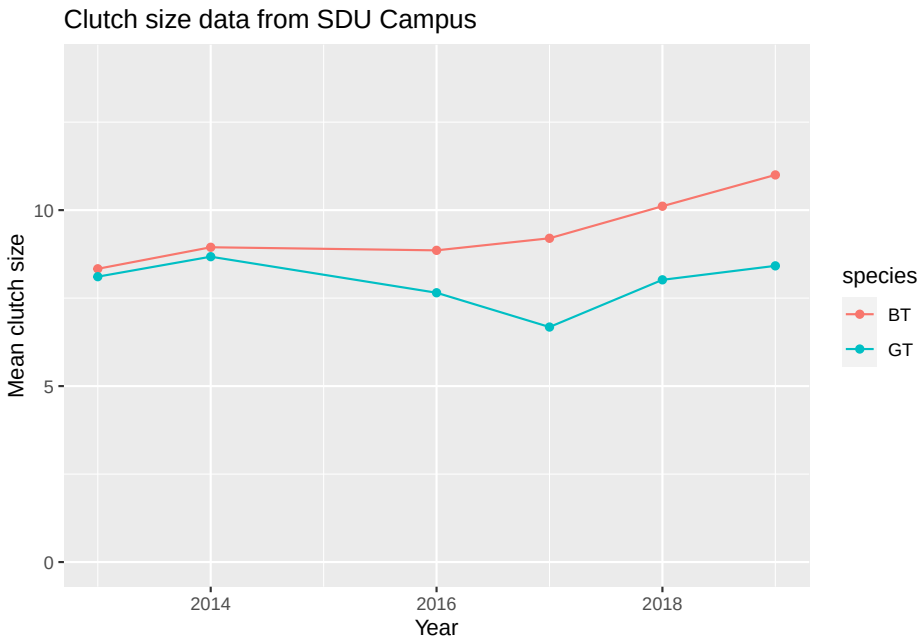
Now I can do the plot again. The only difference to the command is that I need to tell R that I want to colour the lines by species (`colour = species`).

```
ggplot(meanClutch, aes(
  x = Year, y = meanClutchSize,
  colour = species
)) +
  geom_line()
```



I can improve on this by (1) changing the y axis limits (using `ylim`) so that it goes through the full range of my data (0 - 14); (2) adding points (using a `geom_point` layer) where my actual data values are; (3) adding a nicely formatted axis label (using `ylab`); adding a title (`ggtitle`)

```
ggplot(meanClutch, aes(
  x = Year, y = meanClutchSize,
  colour = species
)) +
  geom_line() +
  geom_point() +
  ylim(0, 14) +
  ylab("Mean clutch size") +
  ggtitle("Clutch size data from SDU Campus")
```



10.5 Scatter plots

Finally, lets make a scatter plot. The SDU bird data are not suitable for this type of plot so we'll use the data from a few days ago on suburban bird diversity.

```
df <- read.csv("CourseData/suburbanBirds.csv")
```

Take a look at the data to remind ourselves what it looks like

```
head(df)
```

```
##          Name Year HabitatIndex nIndividuals nSpecies
## 1    Alamotos 1946         10.0           48       12
## 2      Ramona 1946          9.5           30       13
## 3      Verona 1947          9.5           38       15
## 4 Valle Vista 1950          9.5           42       11
## 5    La Gonda 1955         11.0           44       13
## 6     Belgian 1956          9.0           27       14
```

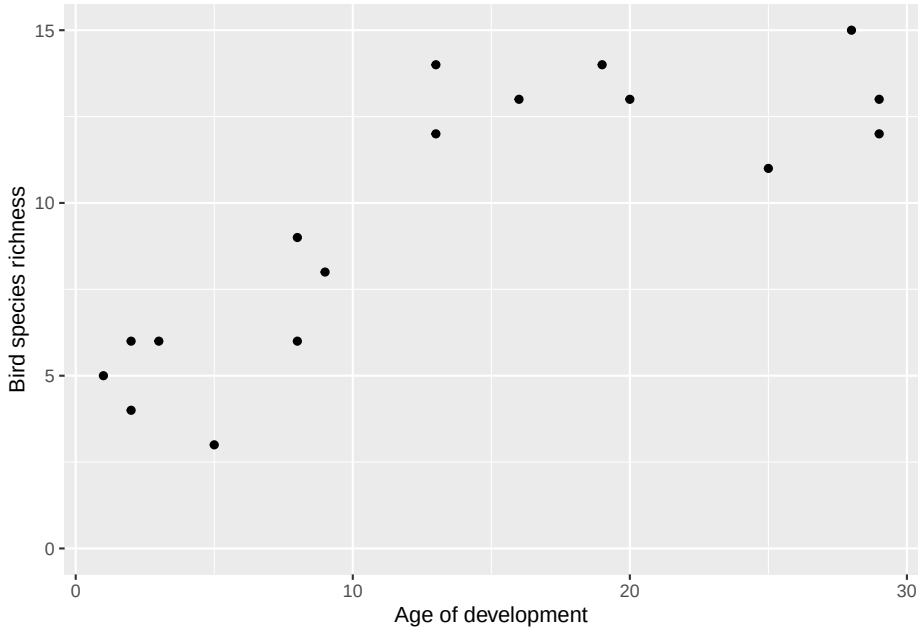
These data show the result of standardised bird surveys at housing developments of different ages in California. The surveys were carried out in 1975, and the data includes the `Year` and number of individual birds seen `nIndividuals` and number of species seen `nSpecies`. The question being addressed is "How does the age of the housing development affect the number of species?"

To investigate this we should first add a new variable for `Age` to the data set. We can do this using the `mutate` function from `dplyr`. This function creates new variables, for example by manipulating existing ones.

```
df <- mutate(df, Age = 1975 - Year)
```

When we have created this variable we can plot the data. For aesthetic reasons I also would like to set the limits on the y-axis to go extend to zero, and I would like to include proper labels on the axes.

```
ggplot(df, aes(x = Age, y = nSpecies)) +
  geom_point() +
  ylim(0, 15) +
  xlab("Age of development") +
  ylab("Bird species richness")
```



This shows very clearly that older developments have more species, but it also appears to show that there is an asymptote around 13 species.

Compare this plot to the one you made with base graphics in a previous class.

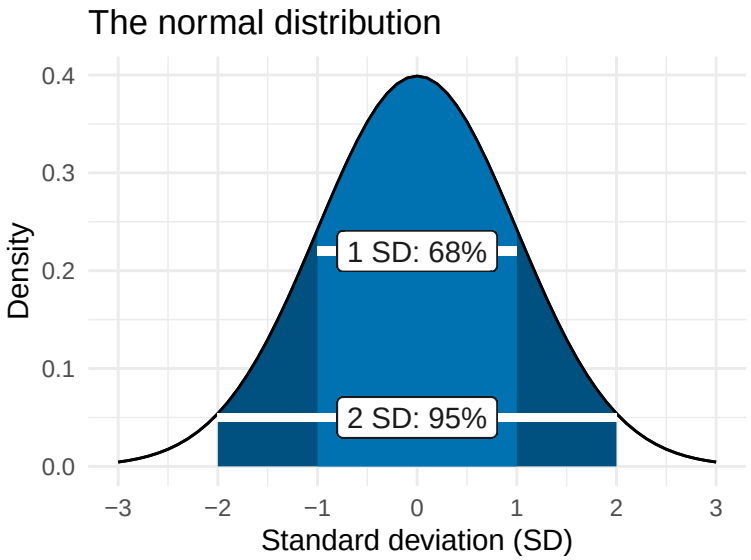
Chapter 11

Distributions and summarising data

This chapter covers two broad topics: the concept of statistical distributions and summarising data. It ends with a brief look at the "law of large numbers".

11.1 Distributions

A statistical distribution is a description of the relative number of times (the *frequency*) possible outcomes will occur if repeated samples were to be taken. They are important because (1) they are useful descriptors of data and (2) they form the basis for assumptions in some statistical approaches. For example, statistical analyses often assume a normal distribution. The normal distribution is symmetrical (centred on the mean) and 68% of observations fall within 1 standard deviation (s.d.), and 95% of observations fall within 2 s.d..



We will use R to simulate some distributions, and explore these to get a feel for them. R has functions for generating random numbers from different kinds of distributions. For example, the function `rnorm` will generate numbers from a normal distribution and `rpois` will generate numbers from a Poisson distribution.

11.2 Normal distribution

The `rnorm` function has three arguments. The first argument is simply the number of values you want to generate. Then, the second and third arguments specify the the mean and standard deviation values of the distribution (i.e. where the distribution is centred and how spread out it is).

The following command will produce 6 numbers from a distribution with a mean value of 5 and a standard deviation of 2.

```
rnorm(6, 5, 2)
```

```
## [1] 0.8860899 5.9511727 0.4674129 6.2696362 3.0969106 5.3058671
```

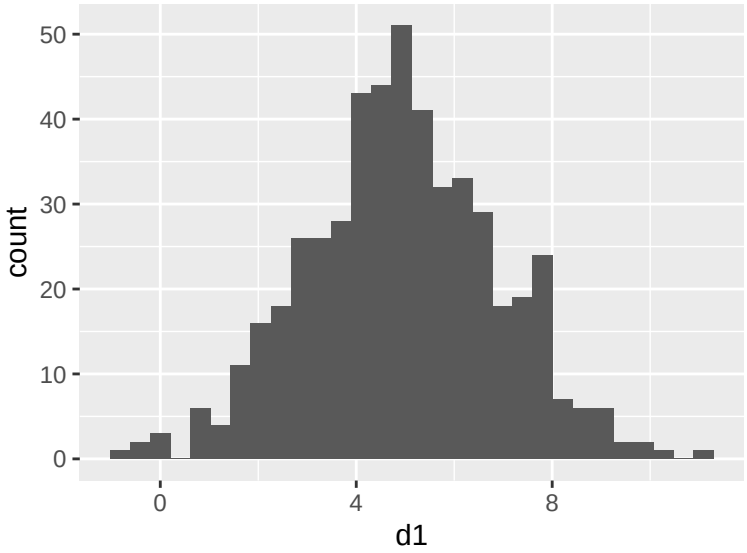
> Try changing the values of the arguments to alter the number of values you generate, and to alter the mean and standard deviation.

Let's use this to generate a larger data frame, and then place markers for the various measures of "spread" onto a plot. *Note that here I put a set of parentheses around the plot code to both display the result AND save the plot as an R object called* `p1`

```
rn <- data.frame(d1 = rnorm(500, 5, 2))
summary(rn) # Take a look
```

```
##        d1
##  Min.   :-0.9862
##  1st Qu.: 3.6789
##  Median : 4.9241
##  Mean   : 4.9399
##  3rd Qu.: 6.2712
##  Max.   :10.9317
```

```
# Plot the data
(p1 <- ggplot(rn, aes(x = d1)) +
  geom_histogram()
)
```



We can calculate the mean and standard deviation using `summarise` (along with other estimates of "spread"). The mean and standard deviation values will be close (but not identical) to the values you set when you generated the distribution.

Note that here I put a set of parentheses around the code to both display the result AND save the result in an object called `sv`

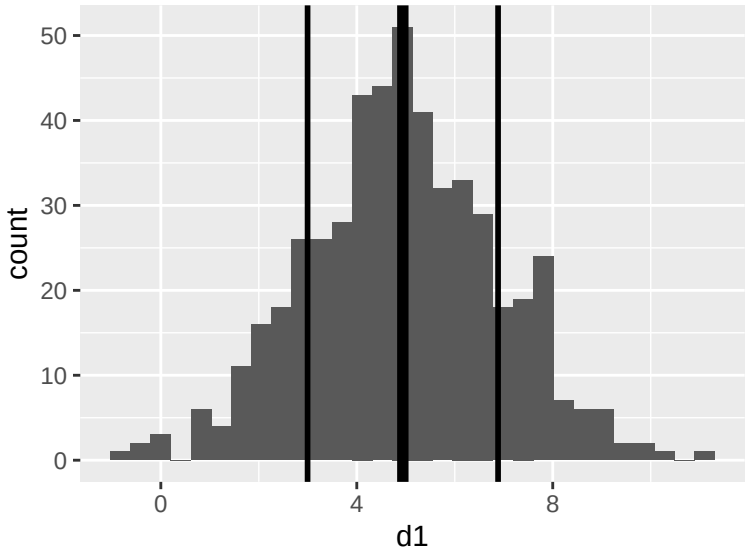
```
(sv <- rn %>%
  summarise(
    meanEst = mean(d1),
```

```
    sdEst = sd(d1),
    varEst = var(d1),
    semEst = sd(d1) / sqrt(n())
  )
)
```

```
##    meanEst    sdEst   varEst     semEst
## 1 4.939908 1.944357 3.780523 0.08695428
```

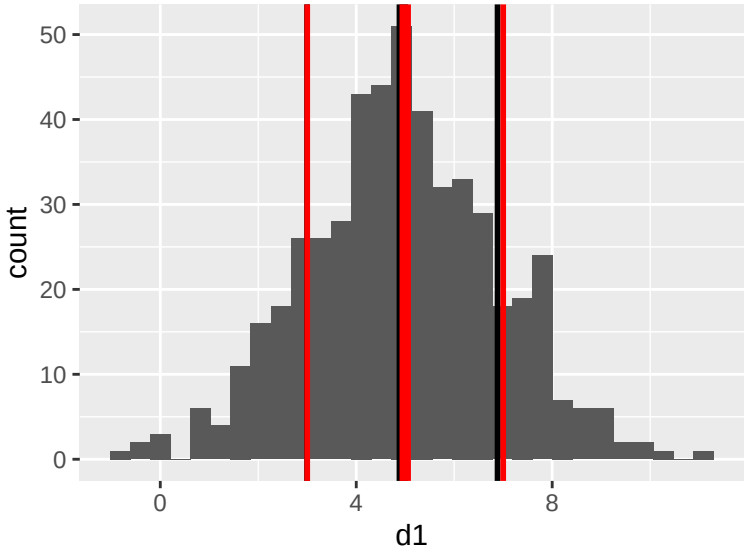
Let's use the function `geom_vline` to add some markers to the plot from above to show these values...

```
(p2 <- p1 +
  geom_vline(xintercept = sv$meanEst, size = 2) + # mean
  geom_vline(xintercept = sv$meanEst + sv$sdEst, size = 1) + # high
  geom_vline(xintercept = sv$meanEst - sv$sdEst, size = 1) # low
)
```



We can compare these with the true values (the values we set when we generated the data), by adding them to the plot in a different colour (mean=5, sd=2).

```
(p3 <- p2 +
  geom_vline(xintercept = 5, size = 2, colour = "red") + # mean
  geom_vline(xintercept = 5 + 2, size = 1, colour = "red") + # high
  geom_vline(xintercept = 5 - 2, size = 1, colour = "red") # low
)
```



Try repeating these plots with data that has different sample sizes. For example, use sample sizes of 5000, 250, 100, 50, 10. What do you notice? You should notice that for smaller sample sizes, the true distribution is not captured very well.

When you calculate the mean and standard deviation, you are actually fitting a simple model: the mean and standard deviation are parameters of the model, which assumes that the data follow a normal distribution.

Try adding lines for the standard error of the mean to one of your histograms.

11.3 Comparing normal distributions

Because normal distributions all have the same shape, it can be hard to grasp the effect of changing the distribution's parameters viewing them in isolation. In this section you will write some code to compare two normal distributions. This approach can be useful when considering whether a proposed experiment will successfully detect a difference between treatment groups. We'll look at this topic, known as "power analysis", in greater detail in a later class. For now we will simply use `ggplot` to get a better feel for the normal distribution.

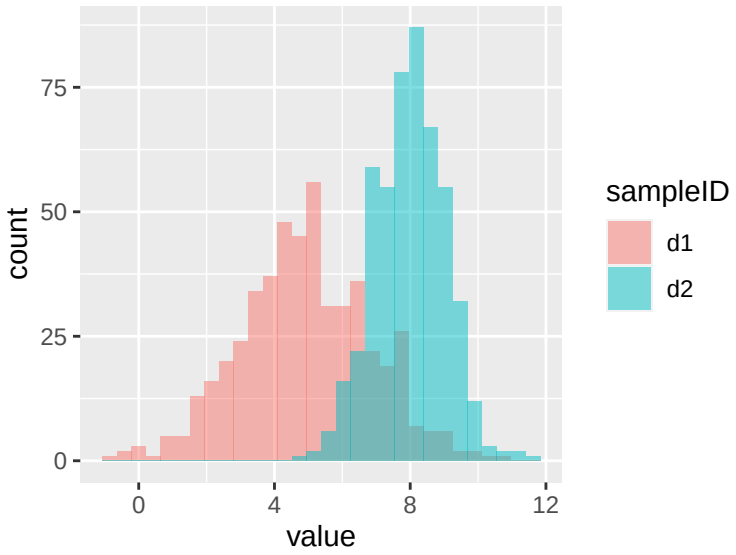
Let's use `rnorm` to generate a larger data frame with two sets of numbers from different distributions: (d1: mean = 5, sd = 2; d2: mean = 8, sd = 1).

```
rn <- data.frame(d1 = rnorm(500, 5, 2), d2 = rnorm(500, 8, 1))
summary(rn)
```

```
##        d1                 d2
##  Min.   :-0.9862   Min.   : 4.628
##  1st Qu.: 3.6789   1st Qu.: 7.305
##  Median : 4.9241   Median : 8.000
##  Mean   : 4.9399   Mean   : 7.978
##  3rd Qu.: 6.2712   3rd Qu.: 8.719
##  Max.   :10.9317   Max.   :11.495
```

The summaries (above) show that the mean and the width of the distributions vary, but we should always plot our data. So lets make a plot in `ggplot`. In the dataset I created I have the data arranged by columns side-by-side, but `ggplot` needs the values to be arranged in a single column, and the identifier of the sample ID in a second column. I can use the function `pivot_longer` to rearrange the data into the required format.

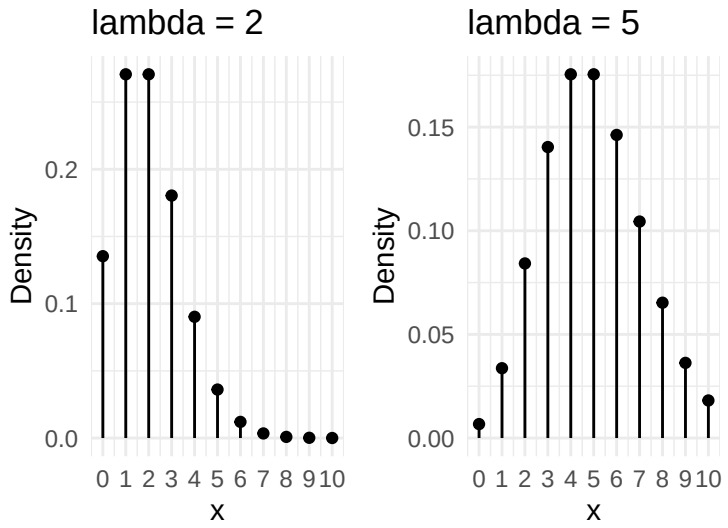
```
rn <- pivot_longer(rn,
  cols = c(d1, d2), names_to = "sampleID",
  values_to = "value"
) # rearrange data

# Plot histograms using "identity", and make them transparent
ggplot(rn, aes(x = value, fill = sampleID)) +
  geom_histogram(position = "identity", alpha = 0.5)
```



Try changing the distributions and re-plotting them (you can change the number of samples, the mean values and the standard deviations).

11.4 Poisson distribution

The Poisson distribution is typically used when dealing with count data. The values must be whole numbers (integers) and they cannot be negative. The shape of the distributions varies with the "`lambda`" parameter. Small values of lambda give more skewed distributions.

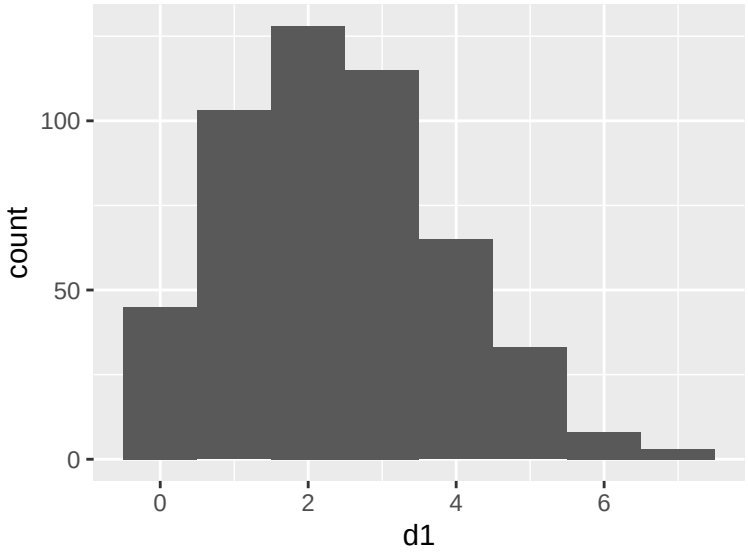


Let's generate and plot some Poisson distributed data.

```
rp <- data.frame(d1 = rpois(500, 2.4))
summary(rp) # Take a look
```

```
##        d1
##  Min.   :0.000
##  1st Qu.:1.000
##  Median :2.000
##  Mean   :2.396
##  3rd Qu.:3.000
##  Max.   :7.000
```

```
# Plot the data
(p1 <- ggplot(rp, aes(x = d1)) +
  geom_histogram(binwidth = 1) # we know the bins will be 1
)
```



Try changing the value of lambda and look at how the shape changes.

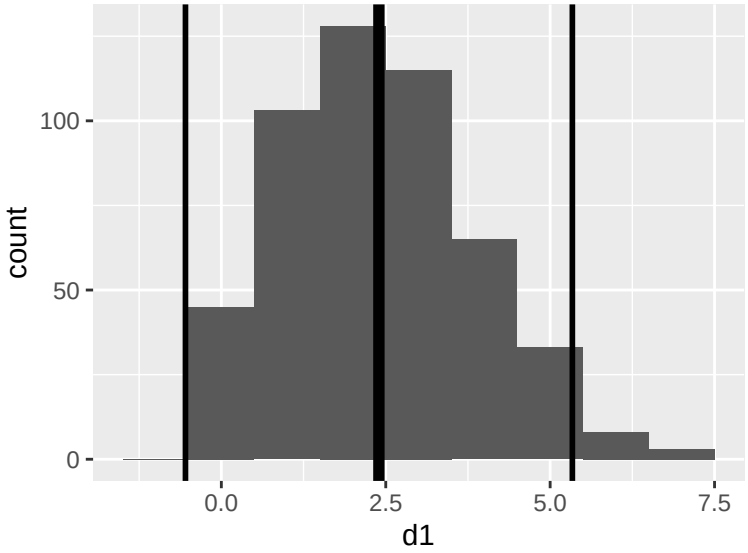
Let's calculate summary statistics of mean and standard deviation for this distribution

```
(sv <- rp %>%
  summarise(
    meanEst = mean(d1),
    sdEst = sd(d1)
  )
)
```

```
##   meanEst    sdEst
## 1   2.396 1.470888
```

Now lets plot the mean and the 2 times the standard deviation on the graph. Remember that for the normal distribution (above) that 95% of the data were within 2 times the standard deviation.

```
p1 +
  geom_vline(xintercept = sv$meanEst, size = 2) +
  geom_vline(xintercept = sv$meanEst + 2 * sv$sdEst, size = 1) +
  geom_vline(xintercept = sv$meanEst - 2 * sv$sdEst, size = 1)
```



This looks like a TERRIBLE fit: The mean is not close to the most common value in the data set and the lower limit of the standard deviation indicates we should expect some negative values - this is impossible for Poisson data. The reason for this is that mean and standard deviation, and therefore standard error, are intended for normally distributed data. When the data come from other distributions we must take another approach.

So how should we summarise this data?

One approach is to report the median as a measure of "central tendency" instead of the mean, and to report "quantiles" of the data along with the range (i.e. minimum and maximum). Quantiles are simply the cut points that divide the data into parts. For example, the 25% quantile is the point where (if the data were arranged in order) one quarter of the values would fall below; the 50% quantile would mark the middle of the data (= the median); the 75% quantile would be the point when three-quarters of the data are below. You can calculate those things using `dplyr`'s `summarise`. However, you can also simply use the base R `summary` command.

```
(sv <- rp %>%
  summarise(
    minVal = min(d1),
    q25 = quantile(d1, 0.25),
    med = median(d1),
    q75 = quantile(d1, 0.75),
    maxVal = max(d1)
  )
)
```

```
##   minVal q25 med q75 maxVal
## 1      0   1   2   3      7
```

```
# base R summary is just as good.
summary(rp$d1)
```

```
##    Min. 1st Qu.  Median    Mean 3rd Qu.    Max.
##   0.000   1.000   2.000   2.396   3.000   7.000
```

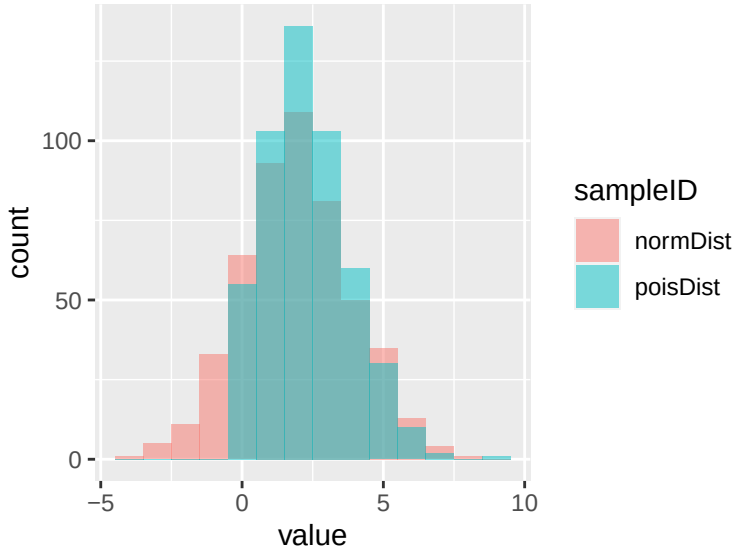
11.5 Comparing normal and Poisson distributions

To get a better feel for how these two distributions differ, lets use the same approach we used above to plot two distributions together.

```
rn <- data.frame(
  normDist = rnorm(500, 2, 2),
  poisDist = rpois(500, 2.4)
)
summary(rn)
```

```
##     normDist          poisDist
##  Min.   :-3.9862   Min.   :0.000
##  1st Qu.: 0.6789   1st Qu.:1.000
##  Median : 1.9241   Median :2.000
##  Mean   : 1.9399   Mean   :2.314
##  3rd Qu.: 3.2712   3rd Qu.:3.000
##  Max.   : 7.9317   Max.   :9.000
```

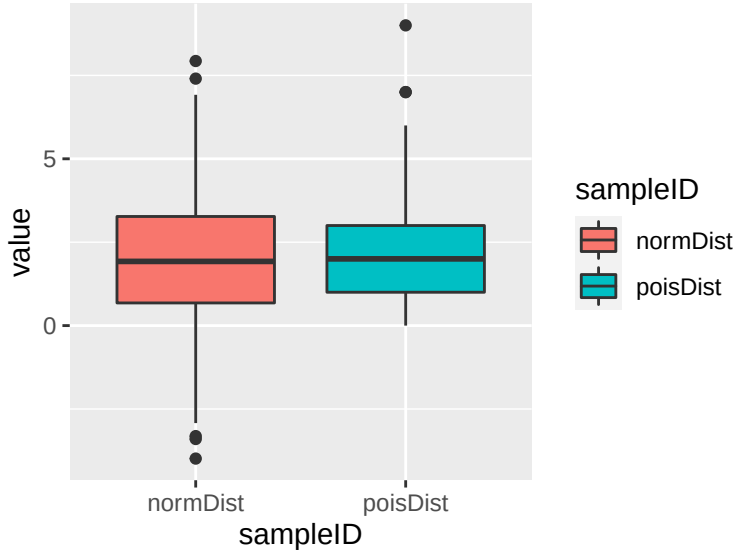
```
rn <- pivot_longer(rn,
  cols = c(normDist, poisDist),
  names_to = "sampleID", values_to = "value"
) # rearrange data

# Plot histograms using "identity", and make them transparent
ggplot(rn, aes(x = value, fill = sampleID)) +
  geom_histogram(position = "identity", alpha = 0.5, binwidth = 1)
```



Try changing the arguments in the `rnorm` and `rpois` commands to change the distributions.

Finally, let's take another view of these data and look at them using box plots. Box plots are a handy alternative to histograms and many people prefer them.

```
ggplot(rn, aes(x = sampleID, y = value, fill = sampleID)) +
  geom_boxplot()
```



You should see the main features of both distributions are captured pretty well. The normal distribution is approximately symmetrical and the Poisson

distribution is skewed (one whisker longer then the other) and cannot be <0. Which graph to you prefer? (there's no right answer!)

11.6 The law of large numbers

The **law of large numbers** is one of the most important ideas in probability. It states that *As sample grows large, the sample mean converges to the population mean.* In other words, as sample size increases, you get a better idea what the true value of the mean is.

In this section you will demonstrate this law using coin tosses or dice throws. Since it is tiresome to toss coins hundreds of times it is convenient to simulate the data using **R**. Conceptually, what we are trying to do here is treat the dice rolling/coin tossing as experiments where the aim is to find the probability of getting a head/tail, or a particular number on the dice. It is useful to use dice and coins because we are pretty sure that we know what the "true" answer is: the probability of throwing a 1 on a fair dice is 1/6, while the probability of throwing a head/tail with a flipped coin is 0.5.

11.6.1 Coin flipping

Here's how to simulate a coin toss in R.

```
coinToss <- c("Heads", "Tails")
sample(coinToss, 1)
```

```
## [1] "Tails"
```

And here is how to simulate 6 coin tosses and make a table of the results. Note, we must use the `replace = TRUE` argument. Please ask if you don't understand why this is necessary.

```
result <- sample(coinToss, 6, replace = TRUE)
table(result)
```

```
## result
## Heads Tails
##     4     2
```

We can "wrap" the `table` function with the `as.data.frame` to turn the data into a data frame that works with `ggplot` You'll probably get different results than me because this is a random process:

```
result <- data.frame(result = sample(coinToss, 6, replace = TRUE))

ggplot(result, aes(x = result)) +
  geom_bar()
```

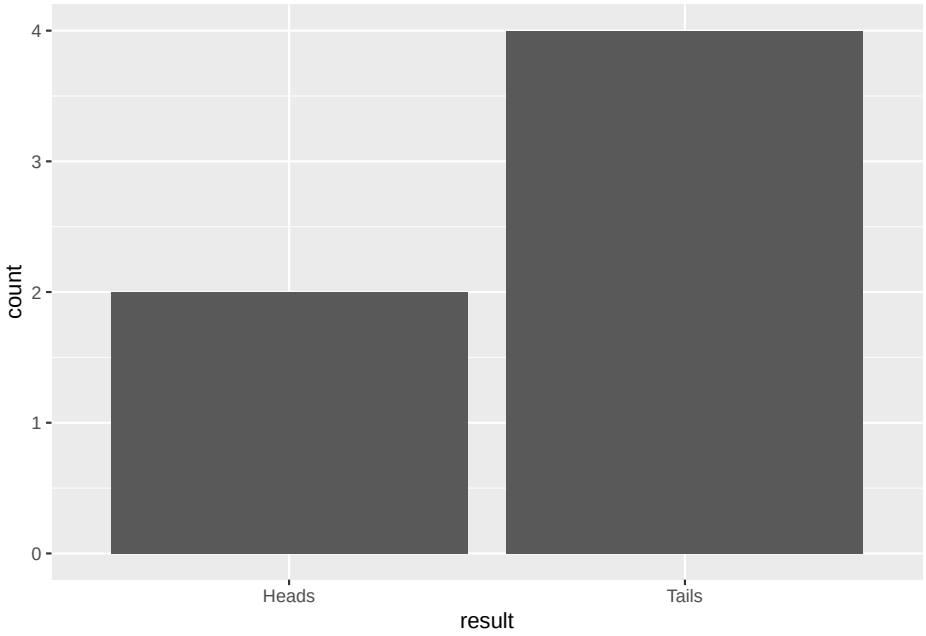


Figure 11.1: Barplot of 6 simulated coin tosses

> Try this several times with small sample sizes (e.g. 4, 6, 8) and see what happens to the proportions of heads/tails. Think about what the expected outcome should be. What do you notice?
> Now increase the sample size (e.g. to 20, 50, 100) and see what happens to the proportions of heads/tails. What do you notice?

11.7 Exercise: Virtual dice

Let's try the same kind of thing with the roll of (virtual) dice.

Here's how to do one roll of the dice:

```
diceRoll <- 1:6
sample(diceRoll, 1)
```

```
[1] 3
```

1) Simulate 10 rolls of the dice, and make a table of results.

2) Now simulate 90 rolls of the dice, and plot the results as a bar plot using `geom_bar` in `ggplot`. Add a horizontal line using `geom_abline` to show the **expected** result based on what you know about probability.

3) Try adjusting the code to simulate dice rolls with small (say, 30) and large (say, 600, or 6000, or 9000) samples. Observe what happens to the proportions, and compare them to the expected value. What does this tell you about the importance of sample size when trying to estimate real phenomena?

Chapter 12

Pimping your plots

In this chapter you will learn, by following examples, how to customise plots made with `ggplot` to improve "readability", or just for aesthetic reasons.

We will cover the following:

1. Modifying axes (log transform, different tick marks/ranges etc.).
2. Colour schemes.
3. *Themes* - built-in sets of styles.
4. Multiple sub-plots in a plot.
5. Saving your plots.

For these examples I will use the dataset on animal life history, `Anage`.

```
x <- read.csv("CourseData/anage_data.csv")
```

You can remind yourself what this data looks like using commands like `summary`, `str` and `names`.

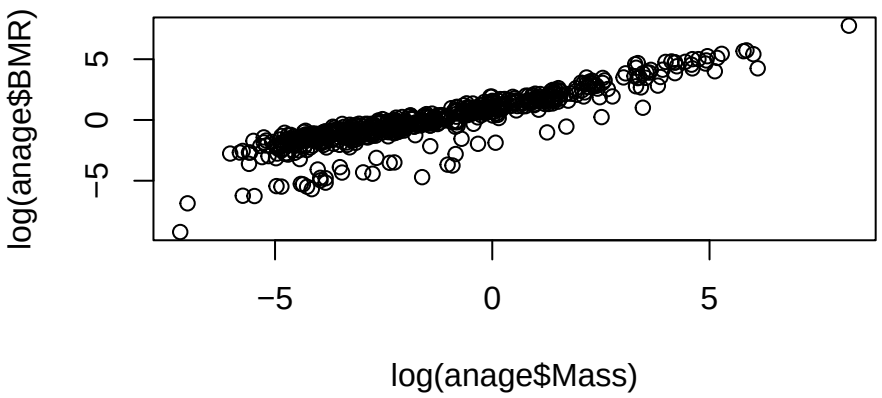
I will process the data a bit to make it easier to work with. One of the commands might be new to you - `rename`. This is simply a way of renaming columns, in this case to make them more "user friendly" (e.g. I want to rename the column "Metabolic.rate..W." to "BMR" (for basal metabolic rate)).

I will also use `mutate` to (1) convert the Mass from grams to kilograms and (2) to make a new variable called "BMRperKg" which standardises metabolic rate by expressing it as rate per kilogram.

```
anage <- x %>%
  mutate(Species = paste(Genus, Species)) %>%
  rename(
    Longevity = "Maximum.longevity..yrs.",
    Mass = "Body.mass..g.",
```

```
    BMR = "Metabolic.rate..W."
  ) %>%
  select(Class, Order, Species, Mass, Longevity, BMR) %>%
  filter(Class %in% c(
    "Aves", "Amphibia",
    "Mammalia", "Reptilia"
  )) %>%
  droplevels() %>%
  # this removes unused "factor levels" e.g. "Insecta"
  mutate(
    Mass = Mass / 1000,
    BMRperKg = BMR / Mass
  )

summary(anage)
```

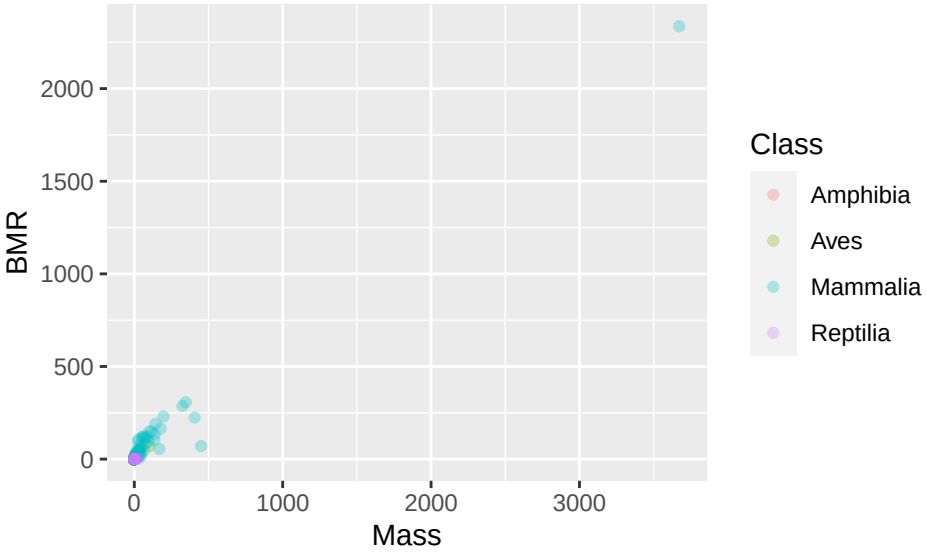
```
##     Class              Order              Species              Mass
##  Length:3231        Length:3231        Length:3231        Min.   :   0.001
##  Class :character   Class :character   Class :character   1st Qu.:   0.026
##  Mode  :character   Mode  :character   Mode  :character   Median :   0.131
##                                                           Mean   :  13.188
##                                                           3rd Qu.:   1.111
##                                                           Max.   :3672.000
##                                                           NA's   :2604
##    Longevity           BMR              BMRperKg
##  Min.   :  0.40   Min.   :   0.0001   Min.   : 0.0454
##  1st Qu.: 10.20   1st Qu.:   0.2655   1st Qu.: 2.2191
##  Median : 16.20   Median :   0.7050   Median : 4.5745
##  Mean   : 19.37   Mean   :  11.8309   Mean   : 7.0439
##  3rd Qu.: 24.45   3rd Qu.:   3.1370   3rd Qu.: 9.8686
##  Max.   :211.00   Max.   :2336.5000   Max.   :45.7692
##  NA's   :432      NA's   :2604        NA's   :2604
```



12.1 A basic plot

Now lets start with a basic plot. You will see a warning about removing rows with missing values. This is just a warning to let you know that there are missing (`NA`) values in the data you are plotting.

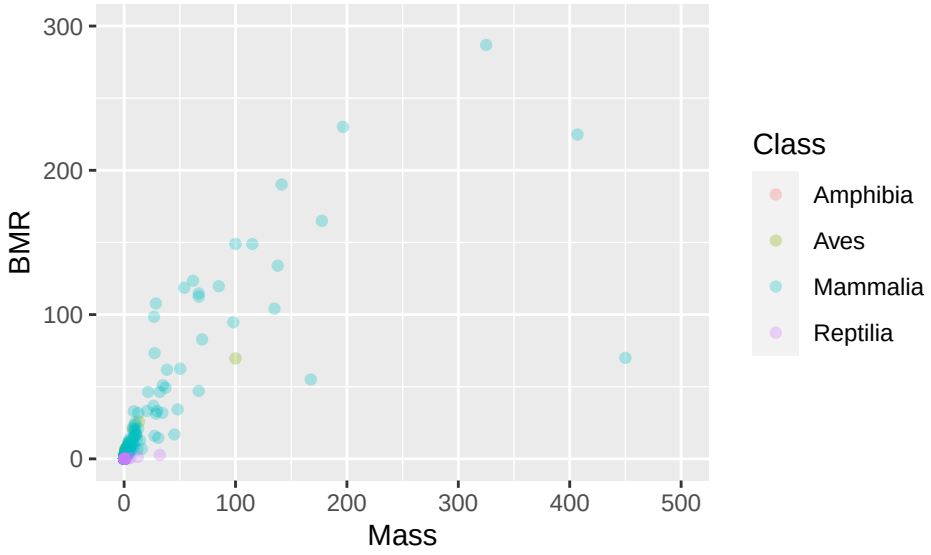
```
(p1 <- ggplot(anage, aes(x = Mass, y = BMR, colour = Class)) +
  geom_point(alpha = 0.3)) # use alpha for transparent points
```



12.2 Axis limits

These points are really spread out. One option to deal with this might be to set the range over which the axes are allowed to go using `xlim` and `ylim`.

```
p1 +
  xlim(0, 500) +
  ylim(0, 300)
```

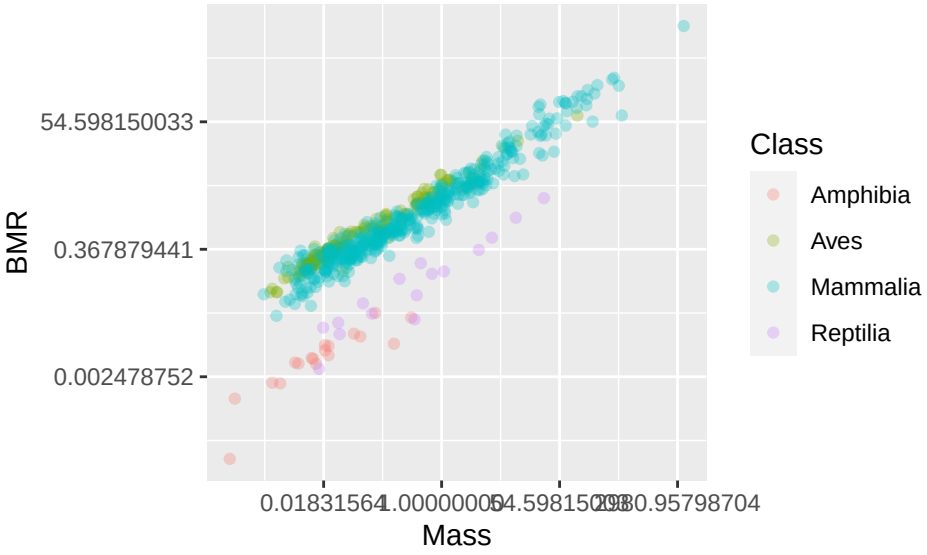


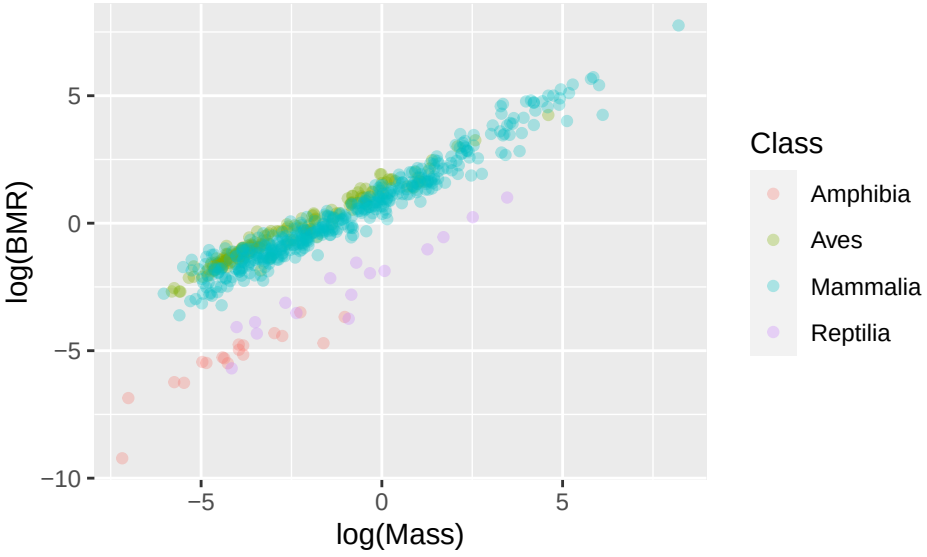
12.3 Transforming the axis (log scale)

In this particular case though use of a log scale would be best because even after focusing on a smaller part of the range of values you can see that the points are still concentrated at smaller values. In a moment, you will also see that log-transforming the data makes the cloud of points pleasingly linear.

You can set a log scale by using the commands `scale_x_continuous(trans = "log")` and `scale_y_continuous(trans = "log")`.

```
(p2 <- p1 +
  scale_x_continuous(trans = "log") +
  scale_y_continuous(trans = "log"))
```

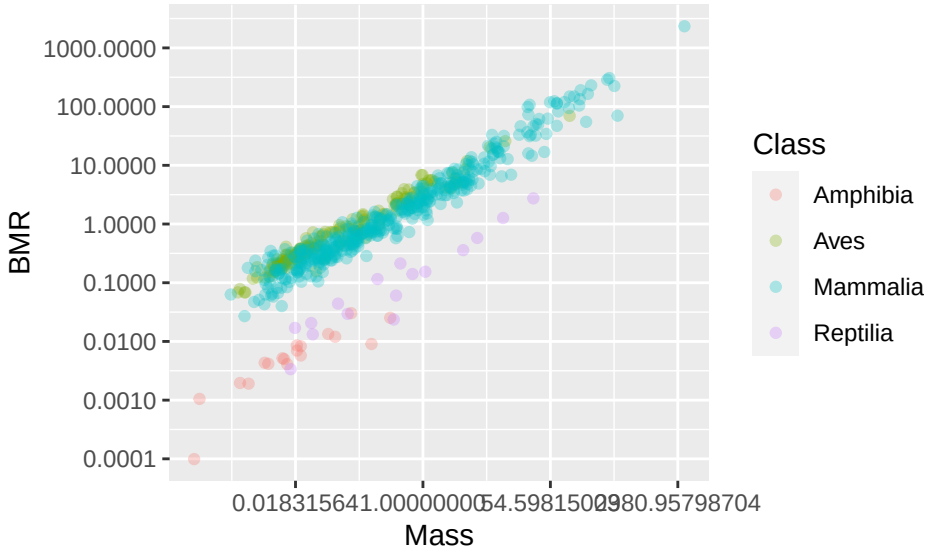




12.4 Changing the axis tick marks

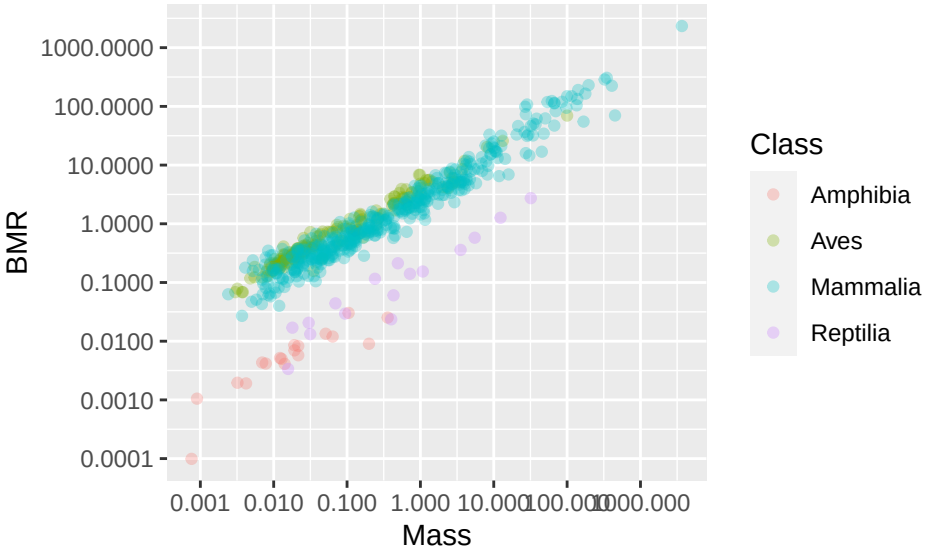
This looks nice. But the numbers on the axis are not very nice. Using `summary(anage$BMR)` tells us that the range of data is from 0.0001 to 2336.5. We could place tick marks anywhere on this axis, but let's try 0.0001, 0.001,0.1, 1,10, 100, 1000.

```
(p2 <- p1 +
  scale_x_continuous(trans = "log") +
  scale_y_continuous(trans = "log", breaks = c(0.0001, 0.001, 0.01, 0.1, 1, 10, 100, 1000)))
```



Using `summary(anage$Mass)` tells us that the range of data is from 0.001 to 3672. We could place tick marks anywhere on this axis, but let's try 0.001,0.1, 1,10, 100, 1000.

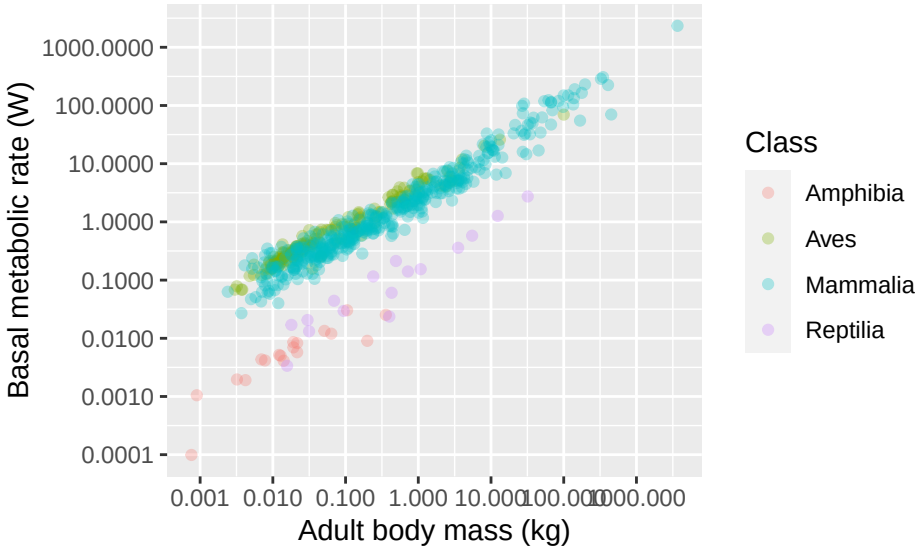
```
(p2 <- p1 +
  scale_x_continuous(
    trans = "log",
    breaks = c(0.001, 0.01, 0.1, 1, 10, 100, 1000)
  ) +
  scale_y_continuous(
    trans = "log",
    breaks = c(0.0001, 0.001, 0.01, 0.1, 1, 10, 100, 1000)
  )
)
```



12.5 Axis labels

Now, let's think about the axis labels. The labels in the plots so far have no units indicated, and might not be easy to interpret for the reader. Let's add units, and also spell out more fully what "BMR" and "Mass" means (the axes is basal metabolic rate in Watts and adult body mass in kg).

```
(p3 <- p2 +
  xlab("Adult body mass (kg)") +
  ylab("Basal metabolic rate (W)")
)
```



12.6 Colours

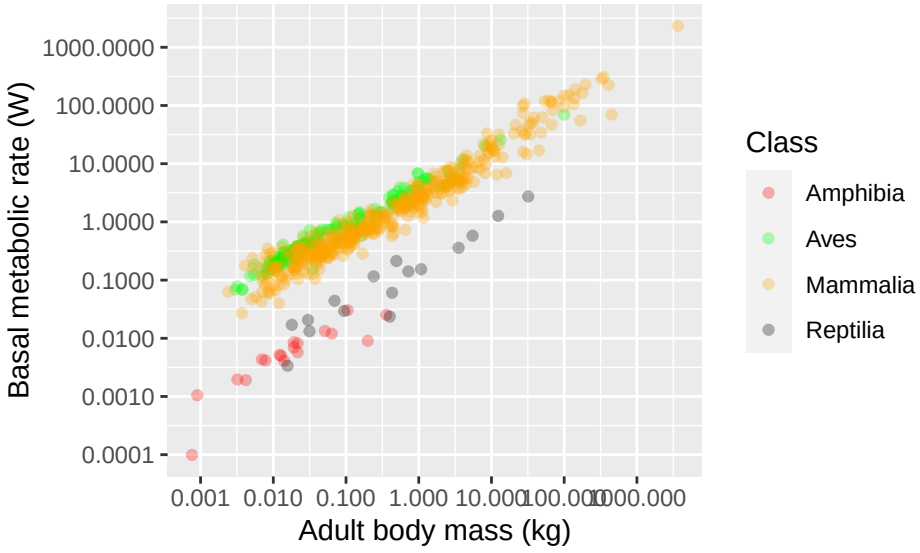
What about those colours? The `ggplot` package uses some default colours that are OK, but sometimes you will want to make a change.

You can "manually" adjust colours using the `scale_colour_manual` function. You can either name individual colours (e.g. "red","green","orange","black")[1], or you can find their so-called "hex-codes" from a site like http://colorbrewer2.org/ or https://htmlcolorcodes.com/color-picker/. You can add a two digit number after the hex code to set the "opaqueness" of the colour. For example "#FF000075" is red, with 75% opacity.

With colour names...

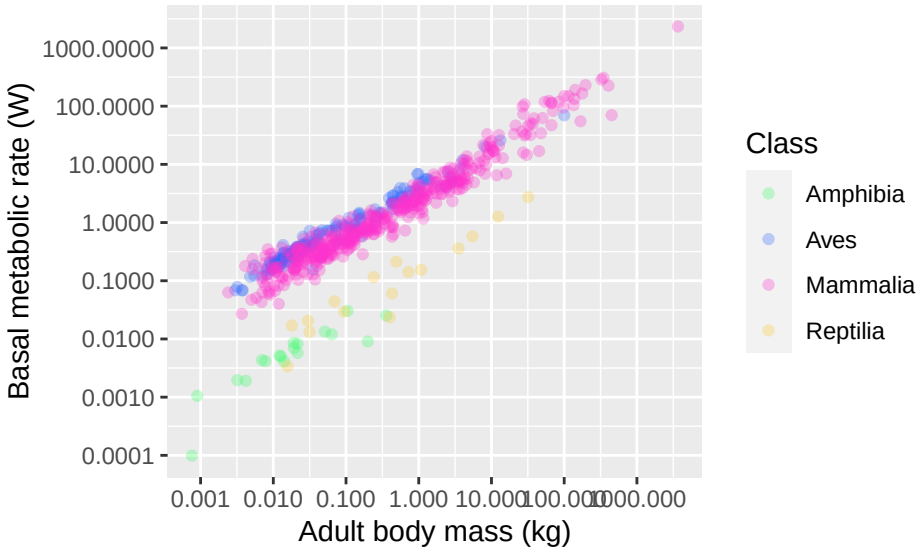
```
p3 +
  scale_colour_manual(values = c(
    "red", "green",
    "orange", "black"
  ))
```

[1] See https://www.r-graph-gallery.com/42-colors-names.html



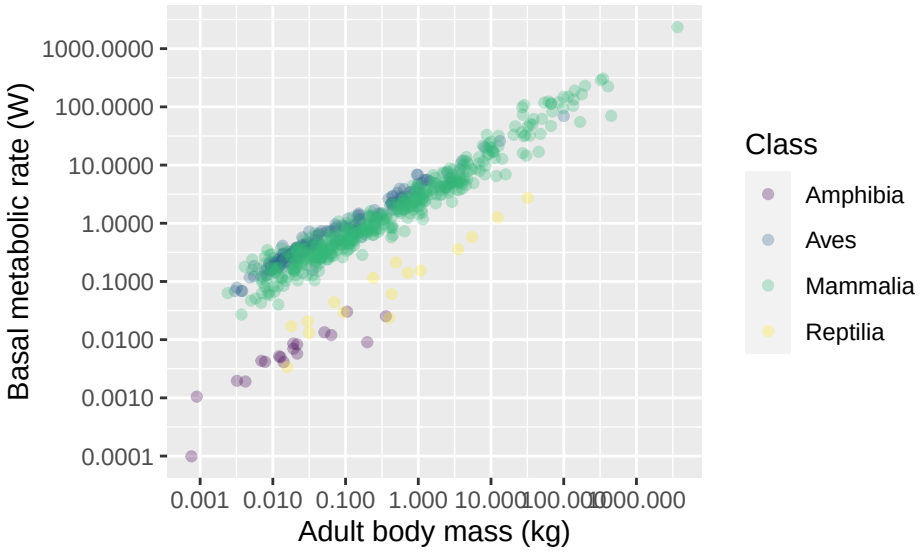
And with some hex codes...

```
p3 +
  scale_colour_manual(values = c(
    "#33FF6475", "#3368FF75", "#FF33CE75",
    "#FFCA3375"
  ))
```



Another alternative is to use some of `ggplot`'s built in "palettes" of colour combinations. For example, there are several palettes called "`viridis`".

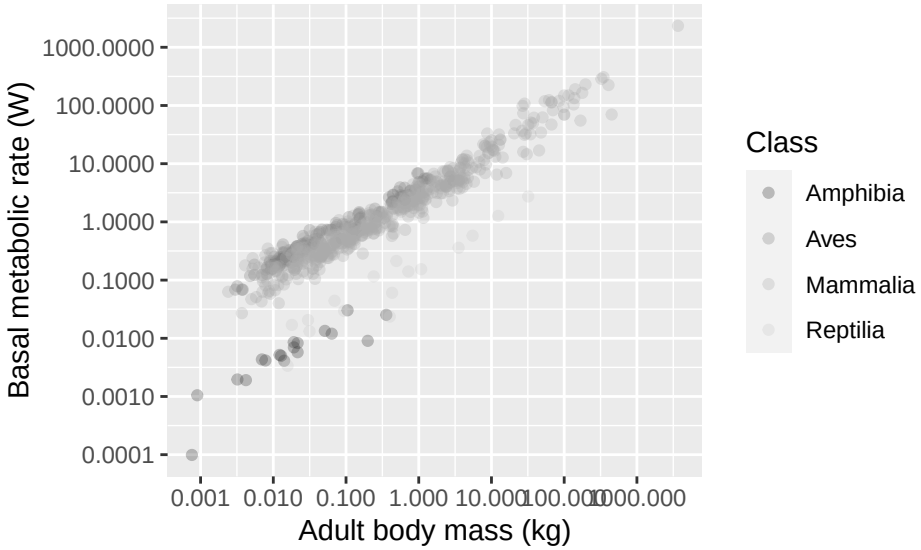
```
p3 +
  scale_colour_viridis_d(option = "D")
```



Try using other `option` arguments `A`, `B`, `C` and `E`. Try also adding an argument for transparency `alpha = 0.5`.

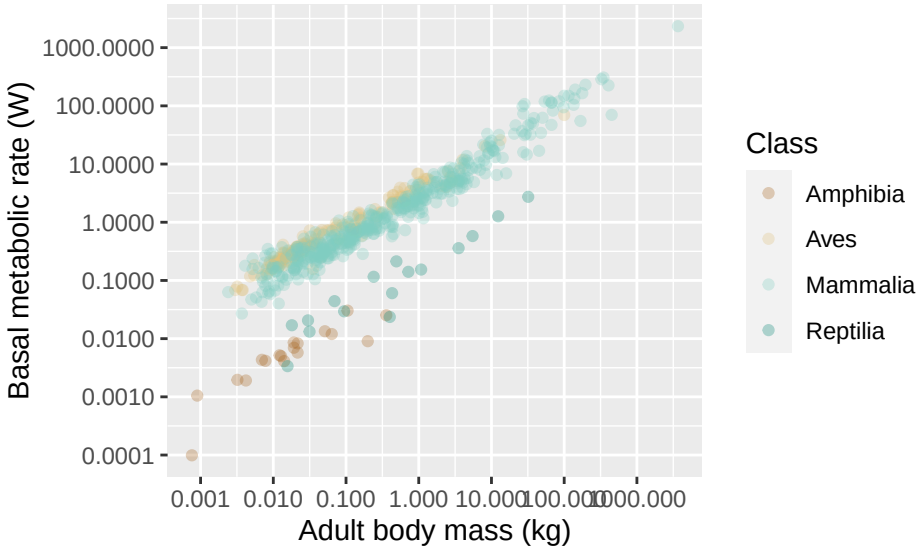
Here's a couple more palettes. There's one for shades of grey…

```
p3 +
  scale_colour_grey()
```



There's another one for various colour schemes, called "colour brewer". Try using "RdGy", "RdYlBu" and "Spectral" see `?scale_colour_brewer` for more options.

```
p3 +
  scale_colour_brewer(palette = "BrBG")
```

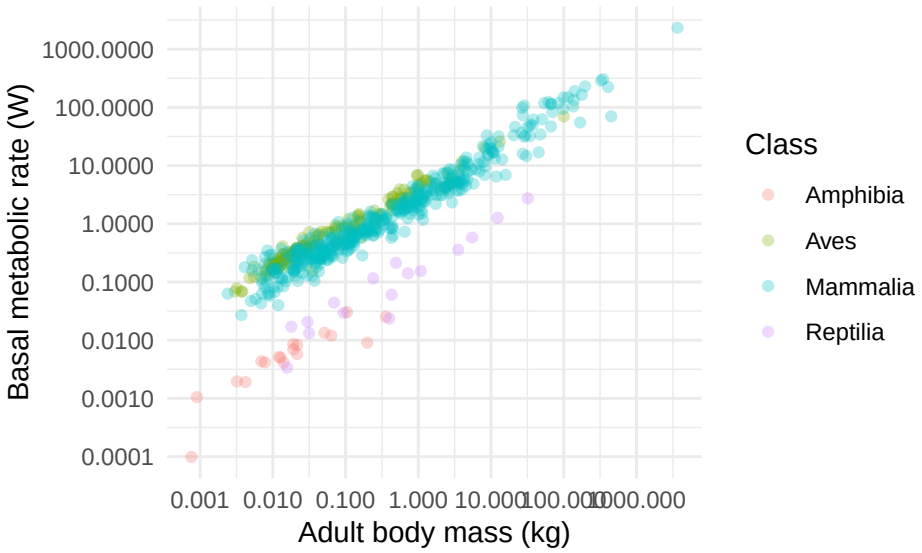


12.7 Themes

Finally, `ggplot` includes the option to set a **theme** for the plots. "Themes"" make adjustments to the "look" of the plot. It is possible to write your own themes, but I recommend to use some ready-made ones. You can implement them by adding them as you would any other addition to the `ggplot` command (e.g. `+ theme_light()`.

There are several themes included with `ggplot`. Try my favourite, `theme_minimal()`. Then try `theme_classic()` and `theme_dark()`.

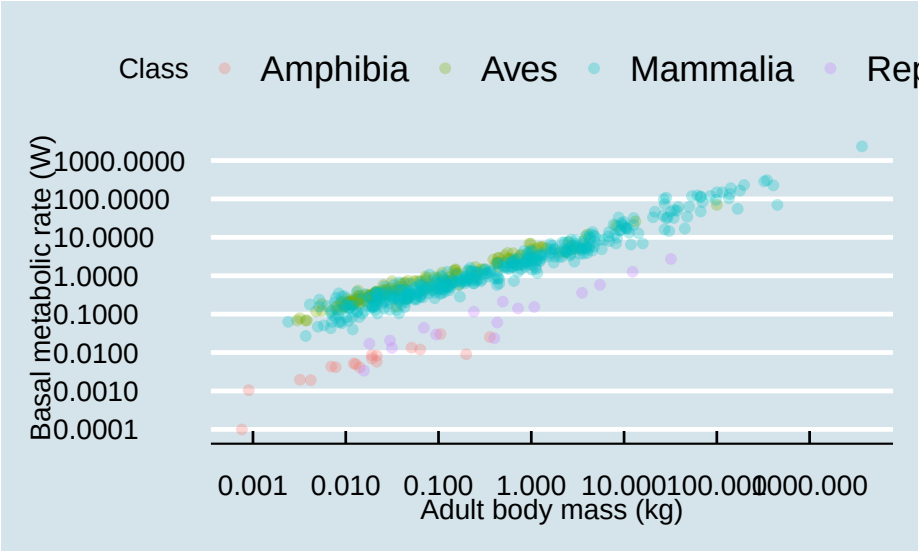
```
(p4 <- p3 +
  theme_minimal()
)
```



For more theme fun, you can install packages that include more themes. The best one is called `ggthemes` (remember that you only need to install the package once). Try `theme_economist()`, `theme_tufte()` and (ugh!) `theme_excel()`. You can see what other themes there in this package at https://jrnold.github.io/ggthemes/reference/index.html (some of them are really ugly in my opinion!).

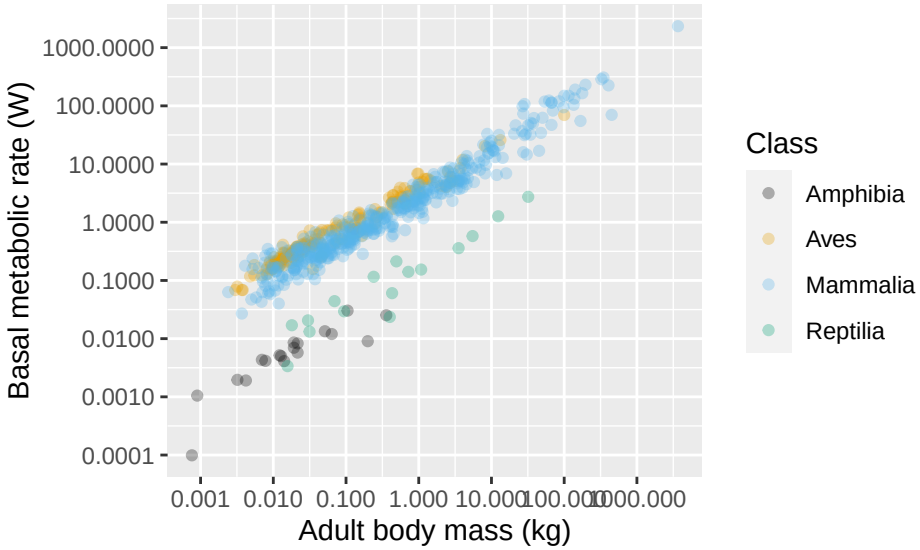
```
install.packages("ggthemes")
```

```
library(ggthemes)
p3 +
  theme_economist()
```



This package also includes some useful colour scales, including some for colour blind people.

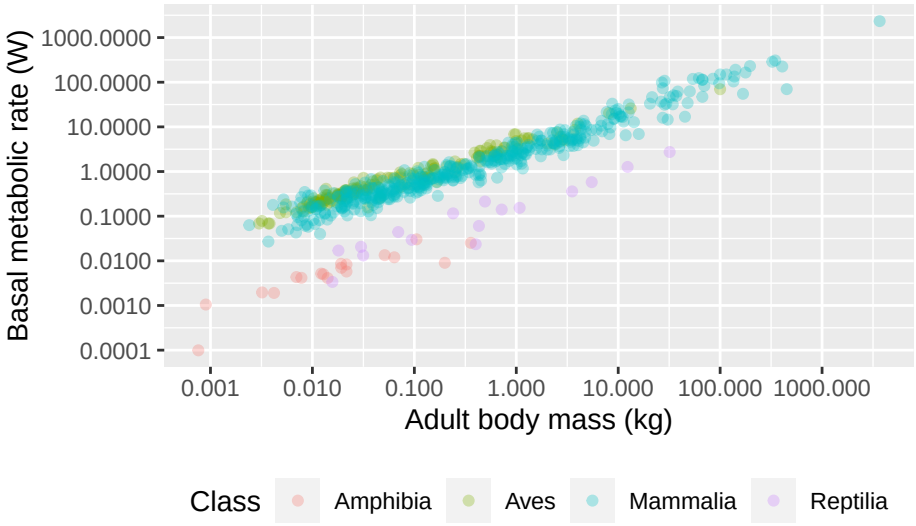
```
p3 +
  scale_color_colorblind()
```



12.8 Moving the legend

By default, the legend is placed on the right. You can move it around by adding a `theme` argument to your plot commands. It can also be placed on the "`top`", "`bottom`", or "`left`". You can also remove the legend altogether by using `legend.position = "none"`. You might also want to remove the legend title using the theme argument `legend.title = element_blank()`.

```
p3 +
  theme(legend.position = "bottom")
```



12.9 Combining multiple plots

It is often useful to combine two or more plots into a single figure. For example, many journals have strict limits on the number of plots so it is useful to combine plots into "Figure 1A and B" etc.

There are several R packages that can do this and my favourite is called `patchwork`.

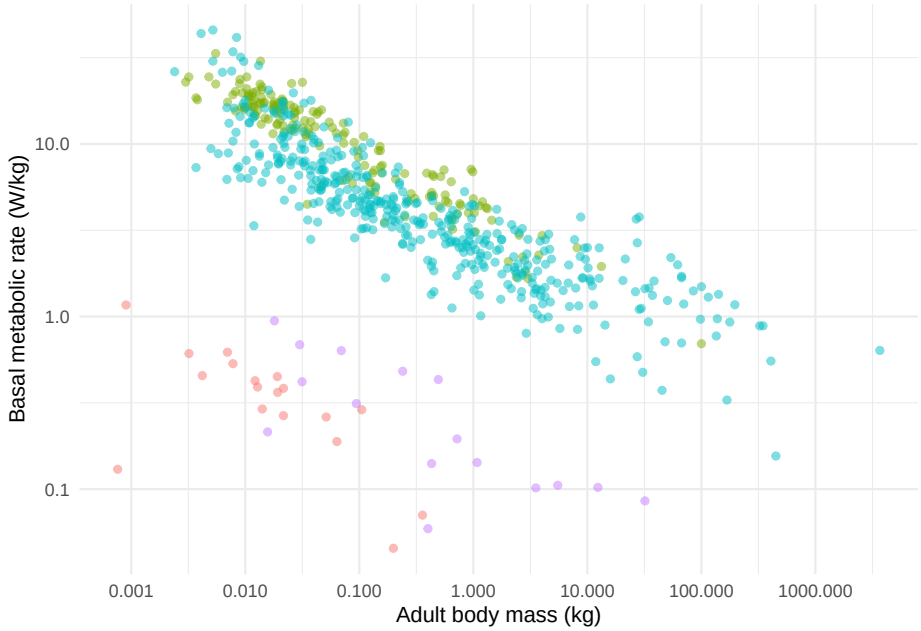
```
install.packages("patchwork") # only need to do this once
```

```
library(patchwork)
```

I will illustrate it by first making another plot, this time showing the relationship between body mass and *standardised* BMR (BMR per kg). Because I am

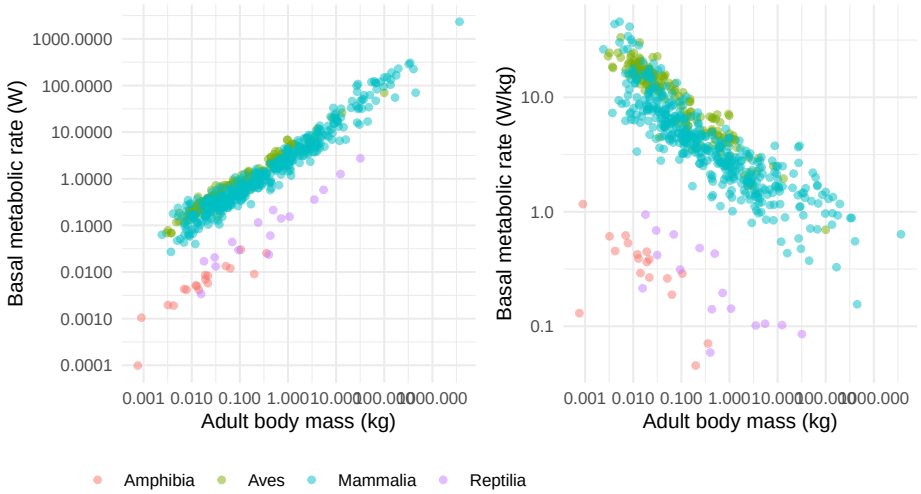
combining the plots into a smaller space I have decided to remove the figure legend (I could put it in the figure caption instead).

```
# PlotA (this is what you have already created above)
plotA <- ggplot(anage, aes(x = Mass, y = BMR, colour = Class)) +
  geom_point(alpha = 0.5) +
  scale_x_continuous(
    trans = "log",
    breaks = c(0.001, 0.01, 0.1, 1, 10, 100, 1000)
  ) +
  scale_y_continuous(
    trans = "log",
    breaks = c(0.0001, 0.001, 0.01, 0.1, 1, 10, 100, 1000)
  ) +
  xlab("Adult body mass (kg)") +
  ylab("Basal metabolic rate (W)") +
  theme_minimal() +
  theme(
    legend.position = "bottom",
    legend.title = element_blank()
  )

(plotB <- ggplot(anage, aes(
  x = Mass, y = BMRperKg,
  colour = Class
)) +
  geom_point(alpha = 0.5) +
  scale_x_continuous(
    trans = "log",
    breaks = c(0.001, 0.01, 0.1, 1, 10, 100, 1000)
  ) +
  scale_y_continuous(
    trans = "log",
    breaks = c(0.0001, 0.001, 0.01, 0.1, 1, 10)
  ) +
  xlab("Adult body mass (kg)") +
  ylab("Basal metabolic rate (W/kg)") +
  theme_minimal() +
  theme(legend.position = "none")
) # This one is wrapped in brackets so that R shows it
```



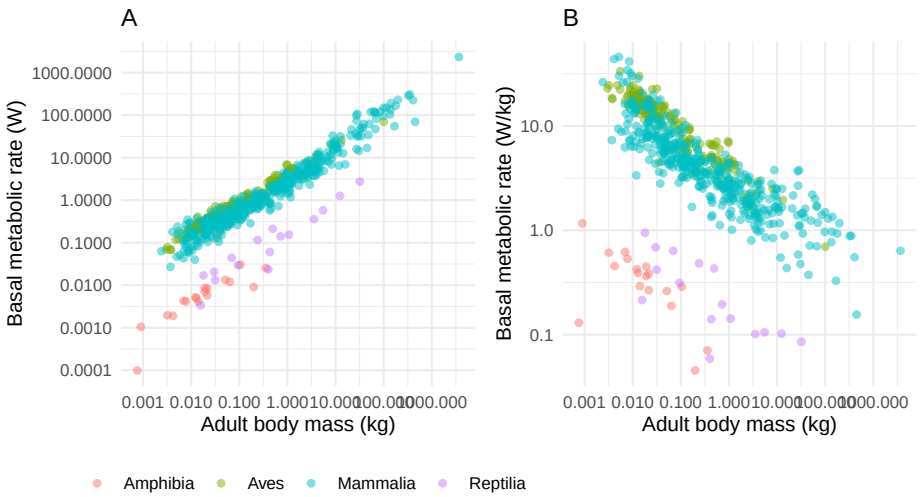
Now I can combine these using the very simple syntax like this:

```
plotA + plotB
```



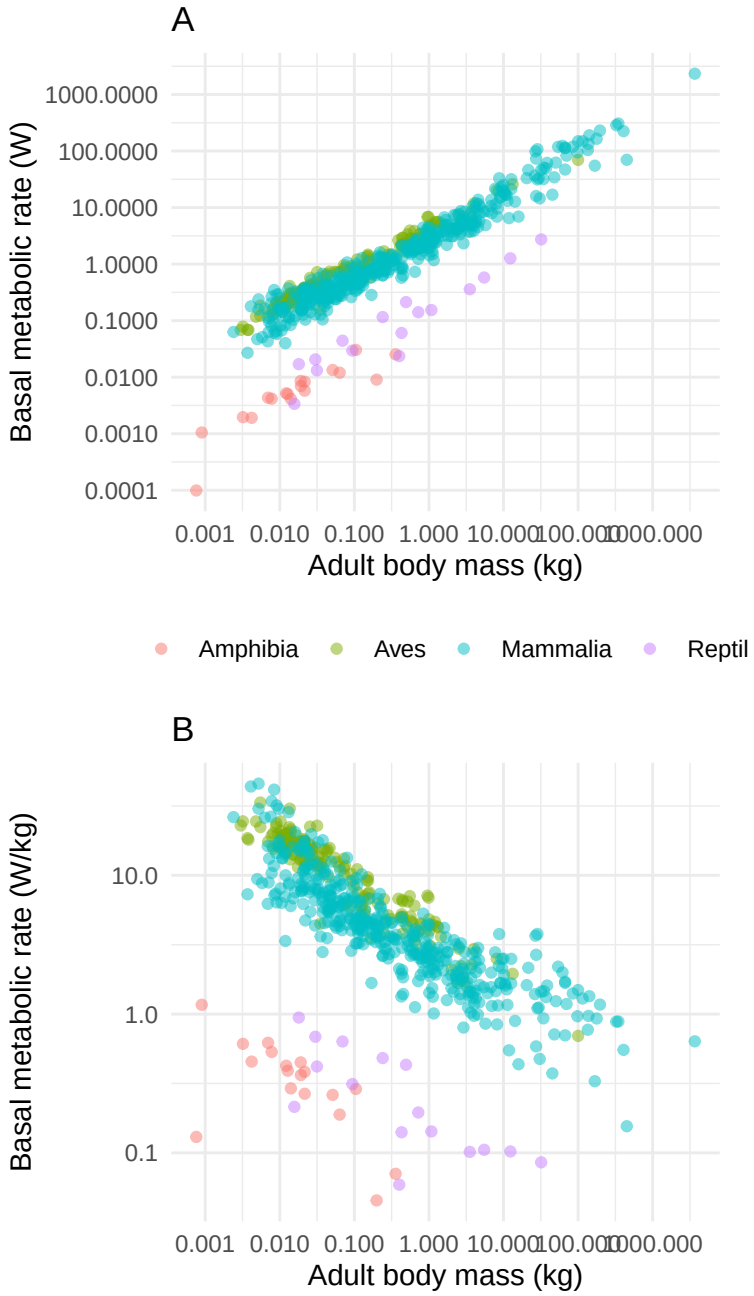
I can add titles using the `ggtitle` command like this.

```
plotA + ggtitle("A") + plotB + ggtitle("B")
```



You could place the sub-plots on top of each other like this.

```
(plotA + ggtitle("A")) / (plotB + ggtitle("B"))
```



12.10 Saving your plot

You should, I think, already know about using the "Export" button in RStudio to save out your plot. This is useful and easy, but you should know that you can also save the plots using a typed command (`ggsave`) in your script. This command is handy because it allows you to automatically set the size, and file name of your plot.

The default setting for `ggsave` is that it will save the last plot that was printed to your computer screen to a file name that you specify. Therefore easiest way to use the command is to simply place the `ggsave` command immediately after your `ggplot` command. You should set the width and height of the plot and the units (the default is inches). It usually takes a few attempts and a bit of trial-and-error to choose the dimensions so that the plot looks nice.

```
ggsave("MySavedPlot1.png", width = 18, height = 10, units = "cm")
```

The command can save to various file types including `png`, `jpeg`, `pdf` (see the `ggplot` help file for more). R knows what file file type is chosen by checking the file extension in the file name (e.g. `.png`). I advise to use `png`.

12.11 Final word on plots

We have covered a lot of ground here. There is a lot to learn, but don't feel like you have to remember all of these commands (I don't). Mostly it is simply a case of remembering that it is *possible* to do these things, and knowing where to look up the commands. Obvious starting points are this course book, and the text book (including the online version!). You can also usually find help by Googling "ggplot" followed by what you are trying to do (e.g. "ggplot change axis ticks"). One of my frequently used web sites is this one http://www.sthda.com/english/ which has an extensive section on `ggplot` (http://www.sthda.com/english/wiki/ggplot2-essentials).

Even though we have covered a lot of ground we have still only gotten a taster of what `ggplot` is capable of. I encourage you to learn more. A useful resource for learning is the online R graph gallery" at https://www.r-graph-gallery.com/, which shows you how to make and modify many types of plot.

Part III

Statistics

Chapter 13

Randomisation Tests

Simple experiments testing for a difference in mean values between two groups usually have the null hypothesis that there is no difference. The alternative hypothesis varies. Sometimes it is simply that the two groups are different (and that the difference could be wither positive or negative). In other cases the alternative hypothesis is that the mean of Group A is less then the mean of Group B (or that it is greater).

Randomisation tests are an intuitive, but computationally intensive way of testing these hypotheses. They have a long history and were first proposed by R.A. Fisher in the 1930s. However they only became convenient when computers became sufficiently fast to do the calculations.

Carrying out a test in R requires that you put your `dplyr` skills to the test. Here you will be guided through an example.

13.1 Randomisation test in R

A new drug has been developed that is supposed to reduce cholesterol levels in men. An experiment has been carried out where 12 human test subjects have been assigned randomly to two groups: "Control" and "Drug". The pharmaceutical company is hoping that the "Drug" group will have lower cholesterol than the "Control" group. The aim here is to do a randomisation test to check that.

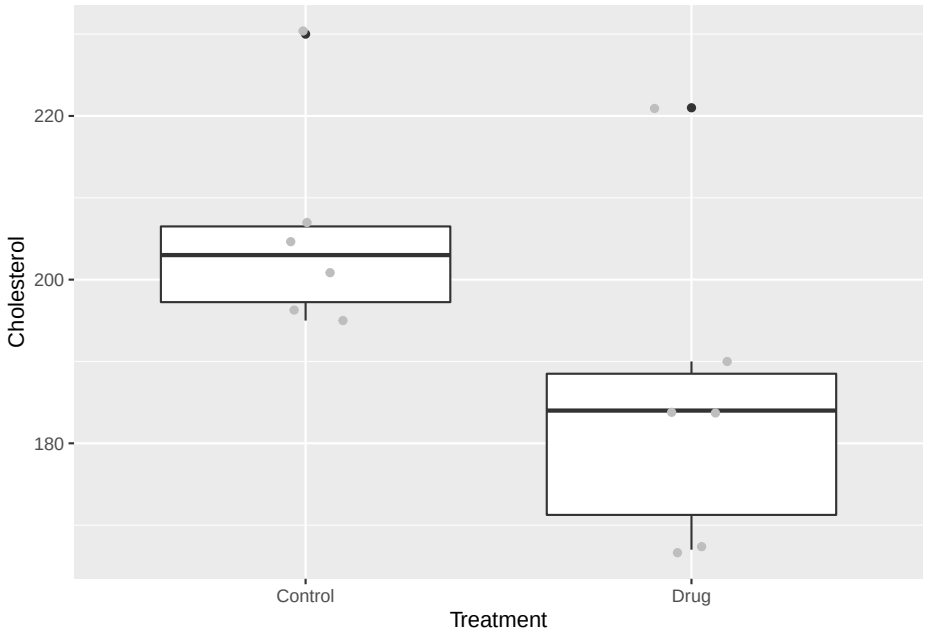
> Remember to load the `dplyr`, `magrittr` and `ggplot` packages, and to set your working directory correctly.

Import the data, called `cholesterol.csv`.

```
ch <- read.csv("CourseData/cholesterol.csv")
```

Let's first take a look at the data by plotting it. I will first plot a boxplot first, and add the jittered points for clarity.

```
ggplot(ch, aes(x = Treatment, y = Cholesterol)) +
  geom_boxplot() +
  geom_jitter(colour = "grey", width = .1)
```



It looks like there might be a difference between the groups. Now let's consider our test statistic and our hypotheses. Our test statistic is the difference in mean cholesterol levels between the two groups: mean of control group minus the mean of the drug group.

The *null hypothesis* is that there is no difference between these two groups (i.e. the difference should be close to 0).

The *alternative hypothesis* is that the mean of the drug group should be less than the mean of the control group.

13.1.1 Calculate the observed difference

There are a few ways of doing this. In base-R you can use the function `tapply` ("table apply"), followed by `diff` ("difference").

```
tapply(ch$Cholesterol, ch$Treatment, mean)
```

```
##  Control     Drug
## 205.6667 185.5000
```

```
diff(tapply(ch$Cholesterol, ch$Treatment, mean))
```

```
##      Drug
## -20.16667
```

Because we are focusing on learning `dplyr`, you can also calculate the means like like this:

```
ch %>% # ch is the cholesterol data
  group_by(Treatment) %>% # group the data by treatment
  summarise(mean = mean(Cholesterol)) # calculate means
```

```
## # A tibble: 2 x 2
##   Treatment  mean
##   <chr>     <dbl>
## 1 Control    206.
## 2 Drug       186.
```

Here the pipes (`%>%`) are passing the result of each function on as input to the next. You can use further commands, `pull` to get the `mean` vector from the summary table, and then use `diff` to calculate the difference between the groups, before passing that to a value called "`observedDiff`".

```
observedDiff <- ch %>%
  group_by(Treatment) %>% # group the data by treatment
  summarise(mean = mean(Cholesterol)) %>% # calculate means
  pull(mean) %>% # extract the mean vector
  diff()
```

This is a complicated set of commands. To make sure that you understand it, try running it bit-by-bit to see what is going on.

13.1.2 Null distribution

Now we ask, what would the world look like if our null hypothesis was *true*. To do this we can disassociate the treatment group variable from the measured cholesterol values. We do this using by using the `mutate` function to replace the `Treatment` variable with a shuffled version of itself with the `sample` function.

Let's try that one time:

```
ch %>%
  mutate(Treatment = sample(Treatment)) %>%
  # shuffle the Treatment data
  group_by(Treatment) %>%
  summarise(mean = mean(Cholesterol)) %>%
  pull(mean) %>%
  diff()
```

```
## [1] -10.16667
```

In this instance, the difference with the shuffled `Treatment` values of -10.167 is rather different from our observed difference of -20.167.

Doing this one time is not much help though - we need to repeat this many times. I suggest that you do it 1000 times here, but some statisticians would suggest 5000 or even 10000 replicates.

We can do this easily in R using the function `replicate` which simply a kind of wrapper that tells R to repeat a command `n` times and then pass the result to a vector. Let's try it first 10 times to see how it works:

```
replicate(
  10,
  ch %>%
    mutate(Treatment = sample(Treatment)) %>%
    group_by(Treatment) %>%
    summarise(mean = mean(Cholesterol)) %>%
    pull(mean) %>%
    diff()
)
```

```
##  [1]  11.500000  -6.500000 -19.166667  -1.833333  -7.833333  21.166667
##  [7] -11.500000 -21.833333  28.833333  -5.833333
```

You can see that the `replicate` command simply does the sampling-recalculation of the mean 10 times.

In the commands below I create 1000 replicates of the shuffled differences. I want to put them in a dataframe to make it easy to plot. Therefore, I first create a `data.frame` called `shuffledData`. This data frame initially has a variable called `rep` which consists of the numbers 1-1000. I then use `mutate` to add the 1000 shuffled differences.

```
shuffledData <- data.frame(rep = 1:1000) %>%
  mutate(shuffledDiffs = replicate(
    1000,
    ch %>%
```

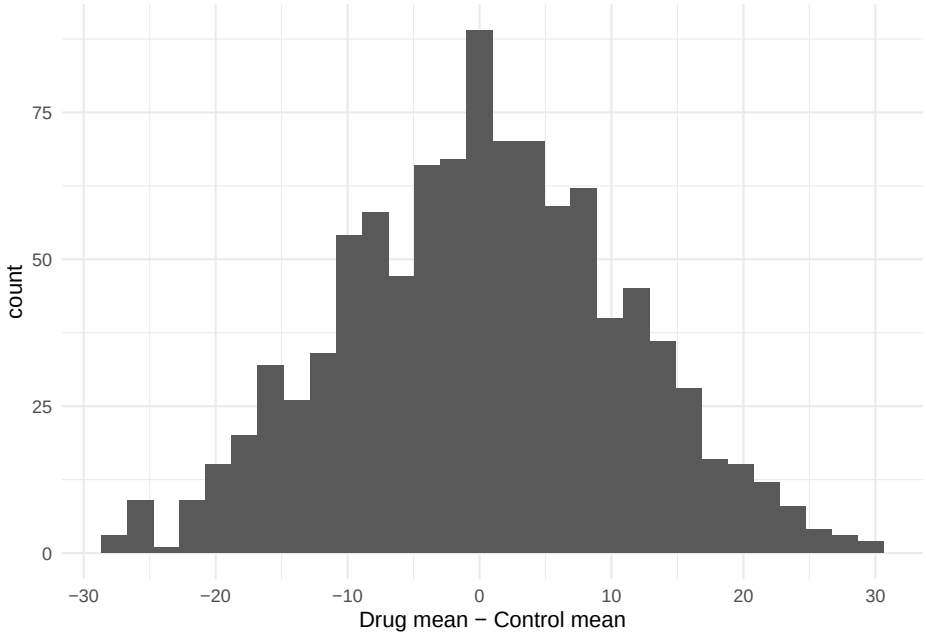
```
    mutate(Treatment = sample(Treatment)) %>%
    group_by(Treatment) %>%
    summarise(mean = mean(Cholesterol)) %>%
    pull(mean) %>%
    diff()
))
```

When you use `summarise`, R will give you a message like this: `summarise() ungrouping output (override with .groups argument)`
This can be annoying, particularly if you are using randomisation tests and summarising hundreds of times. Thankfully, you can turn off this behaviour by setting one of the `dplyr` options like this:
`options(dplyr.summarise.inform = FALSE)`
I suggest to put this code at the beginning of your script if the messages annoy you!

13.1.3 Testing significance

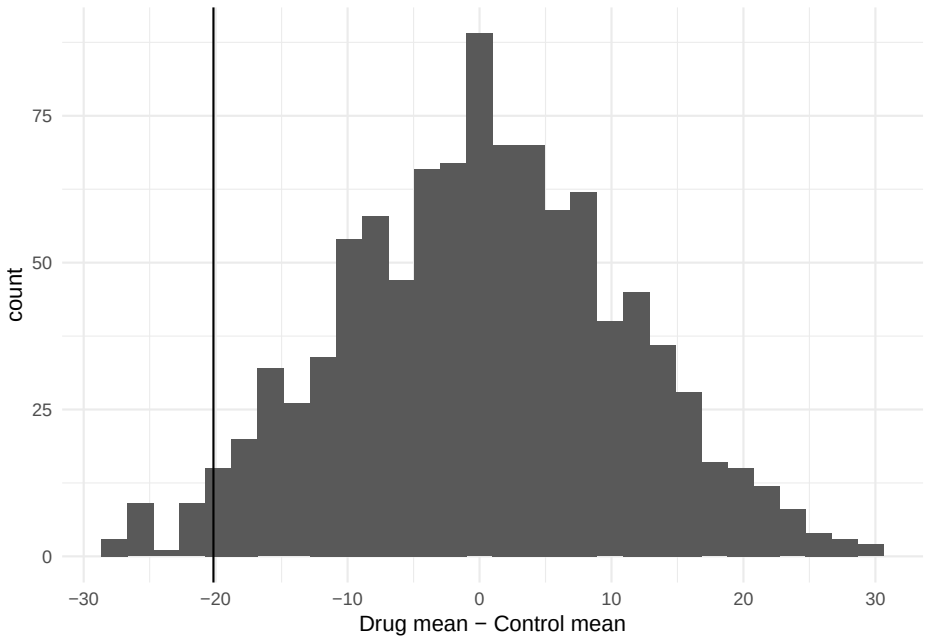
Before formally testing the hypothesis it is useful to visualise what we have created in a histogram. I can use `ggplot` to do this, to create a plot called `p1`. Note that by putting the command in brackets R will both create the plot object, and print it to the screen. Note that because the shuffling of the data is random process your graph will look slightly different to mine.

```
(p1 <- ggplot(shuffledData, aes(x = shuffledDiffs)) +
  geom_histogram() +
  theme_minimal() +
  xlab("Drug mean - Control mean"))
```



You can now add your observed difference (calculated above) to this plot like this:

```
p1 + geom_vline(xintercept = observedDiff)
```



13.1.4 Testing the hypothesis

Recall that the alternative hypothesis is that the observed difference (control mean-drug mean) will be less than 0.

You can see that there are few of the null distribution sample that are as extreme as the observed difference. To calculate a p-value we can simply count these values and express them as a proportion. Note that because the shuffling of the data is random process your result will probably be slightly different to mine.

```
table(shuffledData$shuffledDiffs <= observedDiff)
```

```
##
## FALSE  TRUE
##   977    23
```

So that is 23 of the shuffled values that are equal to or less than the observed difference. The p-value is then simply $23/1000 = 0.023$.

Therefore we can say that the drug appears to be effective at reducing cholesterol.

13.1.5 Writing it up

We can report our findings something like this:

"*To test whether effect of the drug at reducing cholesterol level is statistically significant I did a 1000 replicate randomisation test with the null hypothesis being that there is no difference between the group means and the alternative hypothesis that the mean for the drug treatment is lower than the control treatment. I compared the observed difference to this null distribution to calculate a p-value in a one-sided test.*

The observed mean values of the control and treatment groups 205.667 and 185.500 respectively and the difference between them is therefore **r***-20.167 (drug mean - control mean). Only 25 of the 1000 null distribution replicates were as low or lower than my observed difference value. I conclude that the observed difference between the means of the two treatment groups is statistically significant (p = 0.025)*"

13.2 Paired Randomisation Tests

The paired randomisation test is a one-sample randomisation test where the distribution is tested against a value of 0 (i.e. where there is no difference between the two groups). Often, this distribution is the **difference** in measurements between two sets of measurements taken from the same individuals (or study sites) before and after some treatment has been applied.

I will illustrate this with an example from Everitt (1994) who looked at using cognitive behaviour therapy as a treatment for anorexia. Everitt collected data on weights of people before and after therapy. These data are in the file `anorexiaCBT.csv`

```
# Remember to set your working directory first
an <- read.csv("CourseData/anorexiaCBT.csv")
head(an)
```

```
##   Subject Week01 Week08
## 1       1   80.5   82.2
## 2       2   84.9   85.6
## 3       3   81.5   81.4
## 4       4   82.6   81.9
## 5       5   79.9   76.4
## 6       6   88.7  103.6
```

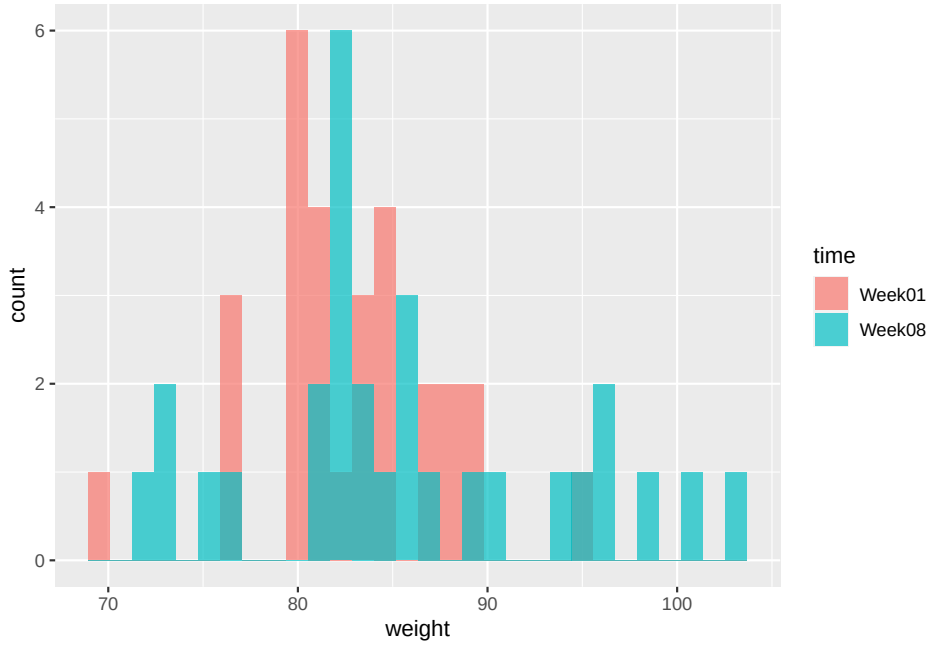
These data are arranged in a so-called "wide" format. To make plotting and analysis data need to be rearranged into a tidy "long" format so that each observation is on a row. We can do this using the `pivot_longer` function:

```
an <- an %>%
  pivot_longer(
    cols = starts_with("Week"), names_to = "time",
    values_to = "weight"
  )
head(an)
```

```
## # A tibble: 6 x 3
##   Subject time   weight
##     <int> <chr>   <dbl>
## 1       1 Week01   80.5
## 2       1 Week08   82.2
## 3       2 Week01   84.9
## 4       2 Week08   85.6
## 5       3 Week01   81.5
## 6       3 Week08   81.4
```

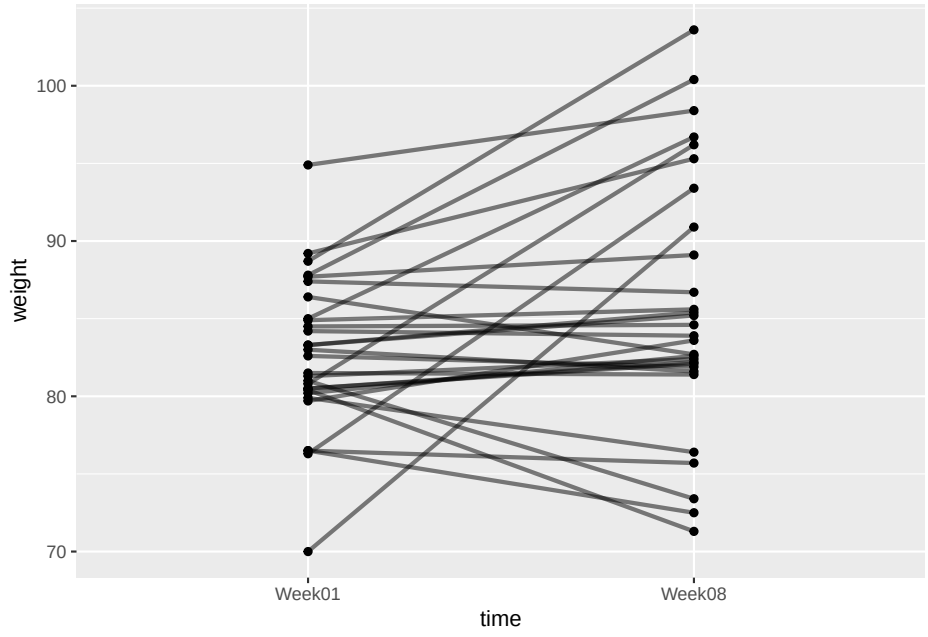
We should *always* plot the data. So here goes.

```
(p1 <- ggplot(an, aes(x = weight, fill = time)) +
  geom_histogram(position = "identity", alpha = .7)
)
```



Another useful way to plot this data is to use an **interaction plot**. In these plots the matched pairs (grouped by `Subject`) are joined together with lines. You can plot one like this:

```
(p2 <- ggplot(an, aes(x = time, y = weight, group = Subject)) +
  geom_point() +
  geom_line(size = 1, alpha = 0.5)
)
```



What we are interested in is whether there has been a change in weight of the subjects after CBT. The null hypothesis is that there is zero change in weight. The alternative hypothesis is that weight has increased.

The starting point for the analysis is to calculate the observed change in weight.

```
an <- an %>%
  group_by(Subject) %>%
  summarise(change = diff(weight))
```

You have created a dataset that looks like this:

```
head(an)
```

```
## # A tibble: 6 x 2
##   Subject change
##     <int>  <dbl>
## 1       1  1.70
## 2       2  0.700
## 3       3 -0.100
## 4       4 -0.700
## 5       5 -3.5
## 6       6 14.9
```

And you can calculate the observed change like this:

```
obsChange <- mean(an$change)
obsChange
```

```
## [1] 3.006897
```

13.2.1 The randomisation test

The logic of this test is that if the experimental treatment has no effect on weight, then the *Before* weight is just as likely to be larger than the *After* weight as it is to be smaller.

Therefore, to carry out this test, we can permute the SIGN of the change in weight (i.e. we randomly flip values from positive to negative and vice versa). We can do this by multiplying by 1 or -1, randomly.

```
head(an)
```

```
## # A tibble: 6 x 2
##   Subject change
##     <int>  <dbl>
## 1       1  1.70
## 2       2  0.700
## 3       3 -0.100
## 4       4 -0.700
## 5       5 -3.5
## 6       6 14.9
```

```
anShuffled <- an %>%
  mutate(sign = sample(c(1, -1),
    size = nrow(an),
    replace = TRUE
  )) %>%
  mutate(shuffledChange = change * sign)
```

Let's take a look at this new shuffled dataset:

```
head(anShuffled)
```

```
## # A tibble: 6 x 4
##   Subject change  sign shuffledChange
##     <int>  <dbl> <dbl>          <dbl>
## 1       1  1.70      1           1.70
## 2       2  0.700     1           0.700
## 3       3 -0.100     1          -0.100
## 4       4 -0.700    -1           0.700
## 5       5 -3.5       1          -3.5
## 6       6 14.9      -1         -14.9
```

We need to calculate the mean of this shuffled vector. We can do this by `pull` to get the vector, and then `mean`.

```
an %>%
  mutate(sign = sample(c(1, -1),
    size = nrow(an),
    replace = TRUE
  )) %>%
  mutate(shuffledChange = change * sign) %>%
  pull(shuffledChange) %>%
  mean()
```

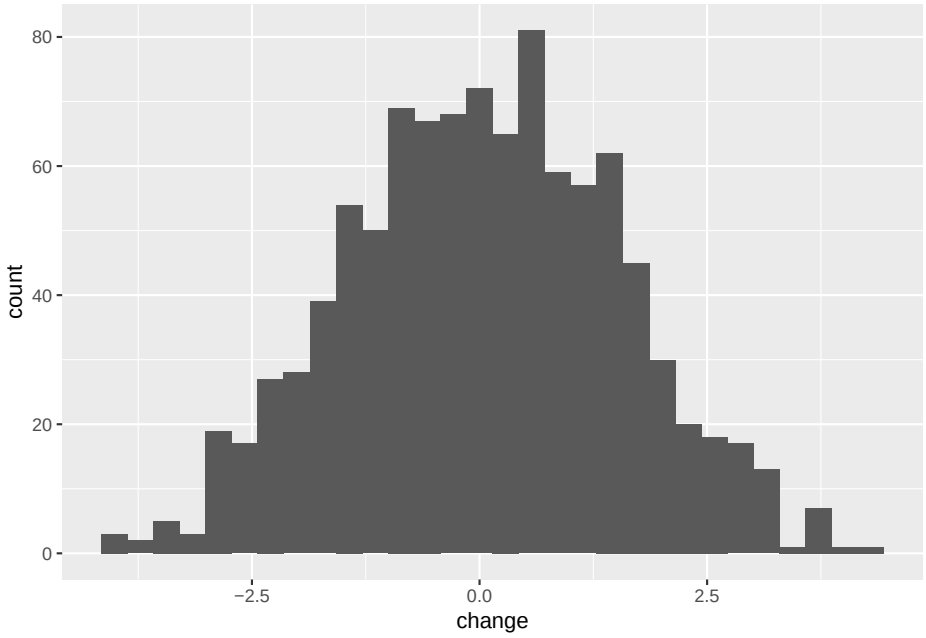
```
## [1] 0.1517241
```

Now we will build a null distribution of changes in weight by repeating this 1000 times. We can do this using the `replicate` function to "wrap" around the function, passing the result into a data frame. We can then compare this null distribution to the observed change.

```
nullDist <- data.frame(
  change =
    replicate(1000, an %>%
      mutate(sign = sample(c(1, -1),
        size = nrow(an),
        replace = TRUE
      )) %>%
      mutate(shuffledChange = change * sign) %>%
      pull(shuffledChange) %>%
      mean())
)
```

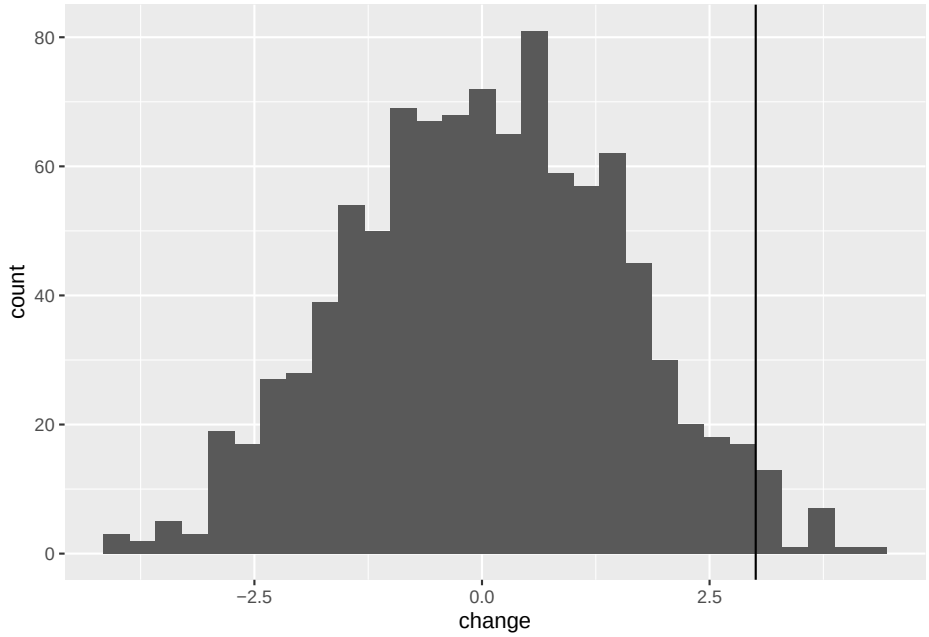
13.2.2 Null distribution

```
(nullDistPlot <- ggplot(nullDist, aes(x = change)) +
  geom_histogram())
```



We can add the observed change as a line to this:

```
nullDistPlot + geom_vline(xintercept = obsChange)
```



13.2.3 The formal hypothesis test

The formal test of significance then works by asking how many of the null distribution replicates are as extreme as the observed change.

```
table(nullDist$change >= obsChange)
```

```
##
## FALSE  TRUE
##   977    23
```

So we can see that 23 of 1000 replicates were greater than or equal to the observed change. This translates to a p-value of 0.023. We can therefore say that the observed change in weight after CBT was significantly greater than what we would expect from chance.

13.3 Exercise: Sexual selection in Hercules beetles

A Hercules beetle is a large rainforest species from South America. Researchers suspect that sexual selection has been operating on the species so that the

males are significantly larger than the females. You are given data[1] on width measurements in cm of a small sample of 20 individuals of each sex. Can you use your skills to report whether males are significantly larger than females.

The data are called `herculesBeetle.csv` and can be found via the course data Dropbox link.

Follow the following prompts to get to your answer:

1. What is your null hypothesis?
2. What is your alternative hypothesis?
3. Import the data.
4. Calculate the mean for each sex (either using `tapply` or using `dplyr` tools)
5. Plot the data as a histogram.
6. Add vertical lines to the plot to indicate the mean values.
7. Now calculate the difference between the mean values using `dplyr` tools, or `tapply`.
8. Use `sample` to randomise the sex column of the data, and recalculate the difference between the mean.
9. Use `replicate` to repeat this 10 times (to ensure that you code works).
10. When your code is working, use `replicate` again, but this time with 1000 replicates and pass the results into a data frame.
11. Use `ggplot` to plot the null distribution you have just created, and add the observed difference.
12. Obtain the p-value for the hypothesis test described above. (1) how many of the shuffled differences are more extreme than the observed distribution (2) what is this expressed as a proportion of the number of replicates.
13. Summarise your result as in a report. Describe the method, followed by the result and conclusion.

[1]This example is from: https://uoftcoders.github.io/rcourse/lec09-Randomization-tests.html

Chapter 14

Comparing two means with a t-test

We will cover the following:

- One-sample t-test
- Paired t-test
- Two-sample t-test ("Welch t-test")

14.1 Some theory

In this theory section I focus on the one-sample t-test, but the concepts apply to the other types of t-test.

The one-sample t-test is used to compare the mean of a sample to some fixed value. For example, we might want to compare pollution levels (e.g. in mg/m^3) in a sample to some acceptable threshold value to help us decide whether we need to take action to prevent or clean up pollution.

One of the assumptions of t-tests (and many other tests/models) is that the distribution of values in the **sample** of data can be described by a normal distribution. If this assumption is true, you can use these data to estimate the parameters of this sample's normal distribution: the mean and standard error of the mean.

The mean gives an estimate of location, and the standard error of the mean (which is calculated as $s/\sqrt{n}$, where s = standard deviation and n = sample size) gives an estimate of precision of this estimate (i.e. how certain is it that the mean value is really where you think it is?)

The t-test then works by comparing your estimated distribution with some fixed value. Sometimes you are asking "is my mean *different* from the value?", other times you are asking "is my mean less than/greater than the value?". This depends on the hypothesis. The default that R-uses is that it tests whether the

mean of your distribution is *different* to the fixed value, but in many cases you should really be framing a **directional** hypothesis.

It is helpful to visualise this, so some examples of the pollution threshold test are shown in the figure below (Figure 14.1). The curves illustrate the estimated normal distributions that describe our estimate of the mean pollution level from some data (e.g. each curve might represent samples from different locations). We are interested in whether the mean values (the vertical dashed lines) are significantly **greater** than the threshold of 100mg/m^2 (solid vertical black line) (this gives us a directional hypothesis).

Formally we do this by establishing two hypotheses a **null hypothesis** and an **alternative hypothesis**. In this case, the null hypothesis is that the mean of the sample measurements is **not** significantly different from the threshold value we define. The alternative hypothesis is that the sample mean is significantly **greater** than this threshold value.

The degree of confidence that we can have that the mean pollution values are different from the threshold value depend on (A) the position of the distribution relative to the threshold value and (B) on the spread of the distribution (the standard deviation/error).

Based on Figure 14.1, which of these four different samples shows a mean value **significantly** greater than 100? (you should be looking at the amount of the normal distribution curve that is overlapping the threshold value.)

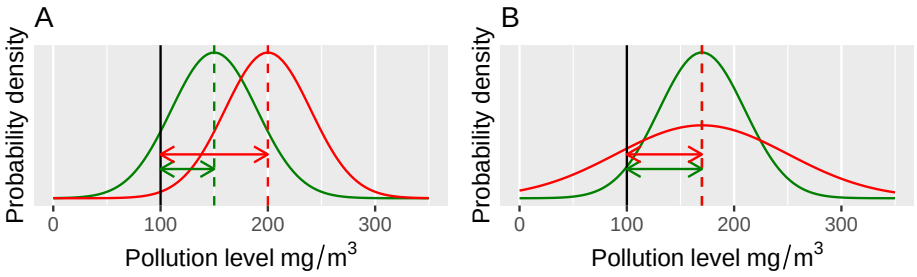


Figure 14.1: Visualisation of a t-test.

This should look familiar – it is the same concept as we used in the class on randomisation tests. If you find it confusing, please go back and review the randomisation test materials!

Another useful way to think about t-tests is that it is a way of distinguishing between **signal** and **noise**: the signal is the mean value of the thing you are measuring, and the noise is the uncertainty in that estimate. This uncertainty could be due to measurement error and/or natural variation. In fact, the *t-value* that the t-test relies on is a ratio between the signal (difference between mean ($\bar{x}$) and threshold (μ_0)) and noise (variability, standard error of the mean ($s/\sqrt{n}$)):

$$t = \frac{\bar{x} - \mu_0}{s/\sqrt{n}}$$

The larger the signal is compared to the noise, the higher the t-value will be. e.g. a t-value of 2 means that the signal was 2 times the variability in the data.

A t-value of zero, or close to zero, means that the signal is "drowned out" by the noise. Therefore, high t-values give you more confidence that the difference is true.

To know if the t-value means that the difference is significant, the t-value is compared to a known theoretical distribution (the t-distribution). The area under the curve of the distribution is 1, but its shape depends on the degrees of freedom (i.e. sample size - 1). The plot below (Figure 14.2) shows three t-distributions of different degrees of freedom (d.f.).

What R is doing when it figures out the p-value is calculating the area under the curve *beyond* the positive/negative values of the t-statistic. If t is small, then this value is large (p-value). If t is large then the area (and the p-value) is small. In the olden-days (>15 years ago) you would have looked these values up in printed tables, but now R does that for us.

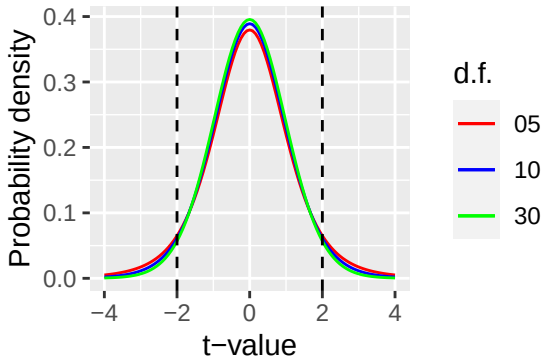


Figure 14.2: The t-distribution

14.2 One sample t-test

Enough theory. Here's how you would apply such a test in R.

> Remember to load the `dplyr`, `magrittr` and `ggplot` packages, and to set your working directory correctly.

Firstly, lets load some data. Because this is a very small example, you can simply cut and paste the data in rather than loading it from a CSV file.

```
pollution <- data.frame(mgm3 = c(
  105, 196, 226, 81, 156, 201, 142, 149, 191, 192,
  178, 185, 231, 76, 207, 138, 146, 175, 114, 155
))
```

First I plot the data (Figure 14.3). One reason for doing this is to check that the data look approximately normally distributed. These data are slightly left-skewed but they are close enough.

```
ggplot(pollution, aes(x = mgm3)) +
  geom_histogram(bins = 8) +
  geom_vline(xintercept = 100)
```

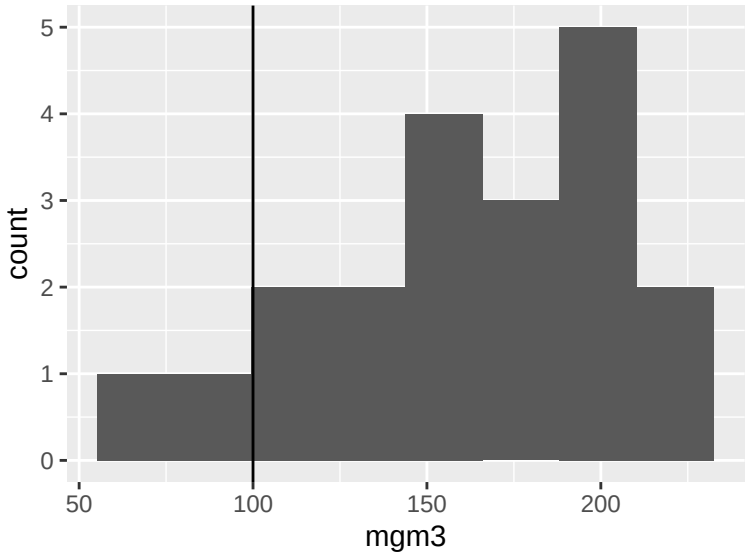


Figure 14.3: Histogram of the pollution data.

Now we can run a t-test in R like this. The command is simple - the first two arguments are the data (`x`) and the the fixed value you are comparing the data to. The final argument defines the alternative hypothesis. This can take values of "`two.sided`", "`less`" or "`greater`" (the default is `two.sided`). In this example, the alternative hypothesis is that the mean of our sample is greater than the threshold of 100.

```
t.test(x = pollution$mgm3, mu = 100, alternative = "greater")
```

```
##
##  One Sample t-test
##
## data:  pollution$mgm3
## t = 6.2824, df = 19, p-value = 0.000002478
## alternative hypothesis: true mean is greater than 100
## 95 percent confidence interval:
##  145.0803      Inf
## sample estimates:
## mean of x
##     162.2
```

The output of the model tells us (1) what type of t-test is being fitted ("`One Sample t-test`"). Then it gives some values for the t-statistic, the degrees of

freedom and the p-value. The model output also tells us that the alternative hypothesis "**true mean is greater than 100**". Because the p-value is very small ($p<0.05$) we can reject the null hypothesis and accept the alternative hypothesis. Finally, the output gives you the confidence interval (the area where we strongly believe the true mean to lie) and the estimate of the mean.

We could report these results like this: *"the mean value of the sample was 162.2 mg/m^3, which is significantly greater than the acceptable threshold of 100 mg/m^3 (t-test: t = 6.2824, df = 19, p-value = 2.478e-06).*

14.3 Doing it "by hand" - where does the t-statistic come from?

At this point, to ensure that you understand where the t-statistic comes from we will calculate the t-statistic using the equation from above. The purpose of this is to illustrate that this is not brain surgery - it all hinges on a straightforward comparison between signal (the difference between mean and threshold in this case) and noise (the variation, or standard error of the mean).

To do this we first need to know the mean value and the threshold (the signal: $\bar{x} - \mu_0$). We can then divide that by the standard error of the mean (the noise: $s/\sqrt{n}$)

Here goes... I first create a vector (`x`) of the values to save typing. Then I show how to calculate `mean` and standard error, before dividing the "signal" by the "noise".

```
# First create a vector of the values
x <- pollution$mgm3

# mean
mean(x)
```

```
## [1] 162.2
```

```
# standard error of the mean
sd(x) / sqrt(length(x))
```

```
## [1] 9.900718
```

```
# Putting it all together
(mean(x) - 100) / (sd(x) / sqrt(length(x)))
```

```
## [1] 6.282373
```

This matches exactly with the t-statistic above!

One can obtain a p-value from a given t-statistic and degrees of freedom like this for a t-test like the one fitted above (the d.f. is the sample size minus one for a one-sample t-test):

```
1 - pt(6.282373, 19)
```

```
## [1] 0.000002477945
```

Again, this matches the value from the `t.test` function above.

14.4 Paired t-test

It's actually quite hard to find examples of one-sample t-tests in biology. In most cases, the one-sample t-tests are really paired t-tests, which are a special case of the one sample test where rather than using the actual values measured, we use the difference between them instead (Figure 14.4).

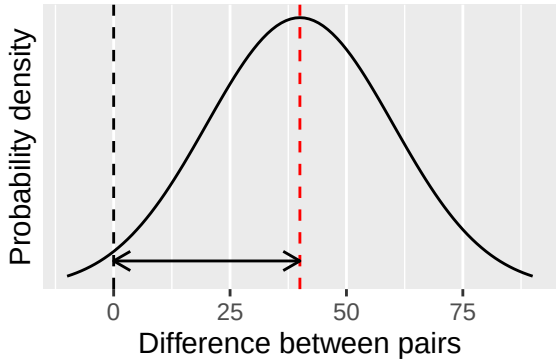


Figure 14.4: Visualising a paired t-test.

Here's a simple example. Anthropologists studied tool use in women from the indigenous Machinguenga of Peru[1]. They estimated the amount of cassava obtained in kg/hour using either a wooden tool (a broken bow) or a metal machete. The study focused on 5 women who were randomly assigned to groups to use the wooden tool then the machete (or vice versa).

The anthropologists hypothesised that using different tools led to different harvesting efficiency. The null hypothesis is that there is no difference between the two groups and that a woman was equally efficient at foraging using either tool. The alternative hypothesis was that there is a difference between the two tools.

[1] A.M. Hurtado, K. Hill (1989). "Experimental Studies of Tool Efficiency Among Machinguenga Women and Implications for Root-Digging Foragers", Journal of Anthropological Research, Vol.45,2,pp207-217.

(NOTE - this could also be formulated as a directional hypothesis e.g. with the expectation that machete is more efficient than the bow.)

First let's import and look at the data. Make sure you understand it. A plot will be fairly useless to tell if the data are normally distributed, so we will simply have to assume that they are. In fact, t-tests are famously robust to non-normality.

```
toolUse <- read.csv("CourseData/toolUse.csv")
toolUse
```

```
##    subjectID    tool amount
## 1          1 machete    119
## 2          1     bow     39
## 3          2 machete    216
## 4          2     bow    114
## 5          3 machete    240
## 6          3     bow    150
## 7          4 machete    129
## 8          4     bow     51
## 9          5 machete    137
## 10         5     bow     60
```

We should now plot our data (Figure 14.5). A nice way of doing this for paired data is to plot points with lines joining the pairs. This way, the slope of the lines is a striking visual indication of the effect.

```
ggplot(toolUse, aes(x = tool, y = amount, group = subjectID)) +
  geom_line() +
  geom_point() +
  xlab("Tool used") +
  ylab("Casava harvested (kg/hr)")
```

We can also look at the mean values and standard deviations:

```
toolUse %>%
  group_by(tool) %>%
  summarise(meanAmount = mean(amount), sdAmount = sd(amount))
```

```
## # A tibble: 2 x 3
##   tool    meanAmount sdAmount
##   <chr>        <dbl>    <dbl>
## 1 bow           82.8     47.3
## 2 machete      168.      55.6
```

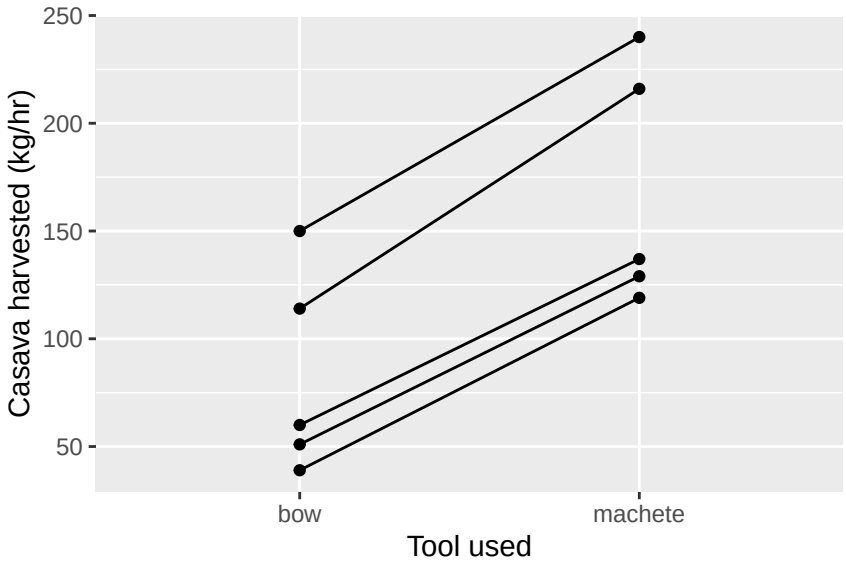


Figure 14.5: An interaction plot for the tool use data

Now lets do a paired t-test to compare the means using the two tools. There are several ways to do a paired t-test. The first is to use a model formula in the command. The formula takes the form `measurements ~ group`. You must also specify the name of the `data.frame` and that the data are paired (`paired = TRUE`).

IMPORTANT: it is very important that the pairs are grouped together in the data frame so that the pairs match up when you filter the data to each group. It is therefore advisable to use `arrange` to sort the data by first the pairing variable (in this case, `subjectID`), and then the explanatory variable (the variable that defines the group - in this case, `tool`.

```
toolUse <- toolUse %>%
  arrange(subjectID, tool)

t.test(amount ~ tool, data = toolUse, paired = TRUE)
```

```
##
##  Paired t-test
##
## data:  amount by tool
## t = -17.98, df = 4, p-value = 0.00005625
## alternative hypothesis: true difference in means is not equal to 0
## 95 percent confidence interval:
##  -98.58738 -72.21262
## sample estimates:
## mean of the differences
##                   -85.4
```

An alternative way is to give the two samples separately in the `t.test` command. To do this you will need to create two vectors containing the data from the two groups like this, using the `dplyr` command `pull` to extract the variable along with `filter` to subset the data:

```
A <- toolUse %>%
  filter(tool == "machete") %>%
  pull(amount)

B <- toolUse %>%
  filter(tool == "bow") %>%
  pull(amount)

t.test(A, B, data = toolUse, paired = TRUE)
```

```
##
##  Paired t-test
##
## data:  A and B
## t = 17.98, df = 4, p-value = 0.00005625
## alternative hypothesis: true difference in means is not equal to 0
## 95 percent confidence interval:
##  72.21262 98.58738
## sample estimates:
## mean of the differences
##                    85.4
```

You would report these results something like this: *"Women harvested cassava more efficiently with a machete (168.2 kg/hr) than with a wooden tool (82.8kg/hr). The difference of 85.4 kg/hr (95% CI 72.2-98.6 kg) was statistically significant (paired t-test: $t = 17.98$, $df = 4$, p-value = 5.625e-05)."*

NOTE: you could add the argument `alternative = "less"` or `greater` to these t-tests to turn them into directional one-tailed hypotheses. However, you should also be aware that the p-value for a one-tailed t-test is always half that of the two-tailed test. Therefore, you could also simply half the p-value when you report it rather than adding the "alternative" argument.

14.5 A paired t-test is a one-sample test.

A paired t-test is the same as a one-sample t-test really. Here's proof.

First we need to calculate the difference between the two measures

```
difference <- A - B
```

Then we can fit the one-sample t-test from above, with the `mu` set as 0 (because the null hypothesis is that there is no difference between the groups). Compare this result with the paired t-test above.

```
t.test(x = difference, mu = 0)
```

```
##
##  One Sample t-test
##
## data:  difference
## t = 17.98, df = 4, p-value = 0.00005625
## alternative hypothesis: true mean is not equal to 0
## 95 percent confidence interval:
##  72.21262 98.58738
## sample estimates:
## mean of x
##      85.4
```

14.6 Two sample t-test

The two sample t-test is used for comparing the means of two samples [no shit!? :)]

You can visualise this by picturing your two distributions (Figure 14.6) and thinking about their overlap. If they overlap a lot the difference between means will not be significant. If they don't overlap very much then the difference between means *will* be significant.

The underlying mathematical machinery for the two-sample t-test is similar to the one-sample and paired t-tests. Again, the important value is the t-statistic, which can be thought of as a measure of signal:noise ratio (*see above*). It is harder to detect a signal (the true difference between means) if there is a lot of noise (the variability, or spread of the distributions), or if the signal is small (the difference between means is small).

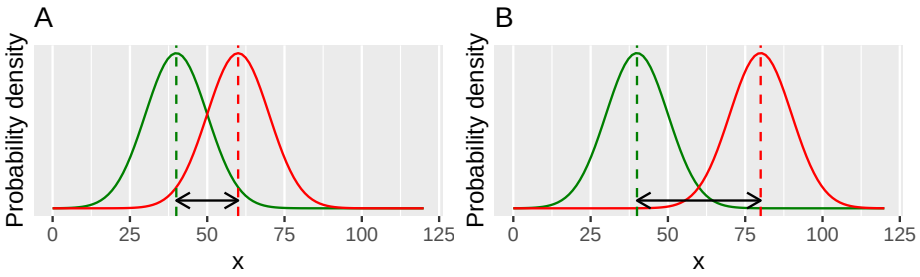


Figure 14.6: A two-sample t-test.

The mathematics involved with calculating the t-statistic is very similar to the one-sample t-test, except the numerator in the fraction is the difference between two means rather than between a mean and a fixed value.

$$t = \frac{\bar{x_1} - \bar{x_2}}{s/\sqrt{n}}$$

So far so good… let's push on and use R to do some statistics.

In this example, we can revisit the class data and ask the question, *Is the reaction time of males different than that of females?* The null hypothesis for this question is that there is no difference in mean reaction times between the two groups. The alternative hypothesis is that there **is** a difference in the mean reaction time between the two groups.

Import the data in the usual way, and subset it to the right year (in the example below I am using 2019 data).

```
x <- read.csv("CourseData/classData.csv") %>% filter(Year == 2019)
```

Then look at the data. Here I do this using a box plot with jittered points (a nice way of plotting data with small sample sizes) (Figure 14.7). From Figure 14.7 it looks like males have a faster reaction time than females, but there is a lot of variation. We need to apply the t-test in a similar way to above.

```
ggplot(x, aes(x = Gender, y = Reaction)) +
  geom_boxplot() +
  geom_jitter(col = "grey60", width = 0.2)
```

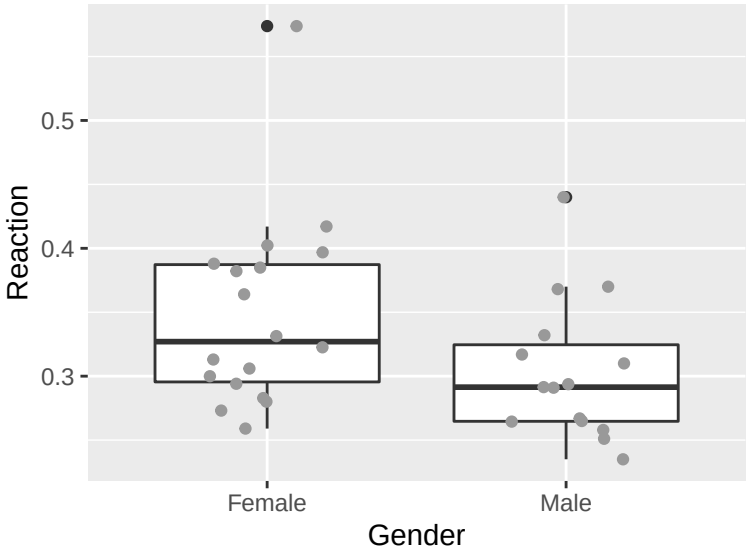


Figure 14.7: Reaction time of both sexes

```
t.test(Reaction ~ Gender, data = x, var.equal = FALSE)
```

```
##
##  Welch Two Sample t-test
##
## data:  Reaction by Gender
## t = 1.9655, df = 30.528, p-value = 0.05851
## alternative hypothesis: true difference in means between group Female and group Ma
## 95 percent confidence interval:
##  -0.001716093  0.091311649
## sample estimates:
## mean in group Female   mean in group Male
##            0.3483778            0.3035800
```

This output first tells us that we are using something called a "Welch Two Sample t-test". This is a form of the two-sample t-test that relaxes the assumption that the variance in the two groups is the same. This is a good thing. Although it *is* possible to fit a t-test with equal variances, I recommend that you stick with the default Welch's test and **not** make this limiting assumption.

Then we are told the t-statistic (1.965), the degrees of freedom (30.528) and the p-value (0.059). We must therefore accept the null hypothesis: there is no significant difference between the two groups. Males are *not* faster than females. We could write report this something like this:

"*Although females had a slightly slower reaction time than males (0.348 seconds compared to 0.304 seconds), this difference was not statistically significant (Welch t-test: t= 1.965, d.f.= 30.528), p=0.059).*"

Note: With a t-test that *did* assume equal variances in the two groups, the d.f. is calculated as the sample size - 2 (the number of groups). You can do this by adding the argument "`var.equal = TRUE`" to the t-test command. With the Welch test, the appropriate degrees of freedom are estimated by looking at the sample sizes and variances in the two groups. The details of this are beyond the scope of this course.

14.7 t-tests are linear models

It is also possible to formulate t-tests as linear models (using the `lm` function). To do this with the paired t-test you would specify a model that estimates an intercept. In R you can do this by writing the formula as `x ~ 1`. So, for the tool use example you can write the code like this:

```
mod1 <- lm(difference ~ 1)
summary(mod1)
```

```
##
## Call:
## lm(formula = difference ~ 1)
##
## Residuals:
##    1    2    3    4    5
## -5.4 16.6  4.6 -7.4 -8.4
##
## Coefficients:
##             Estimate Std. Error t value  Pr(>|t|)
## (Intercept)    85.40       4.75   17.98 0.0000562 ***
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## Residual standard error: 10.62 on 4 degrees of freedom
```

If you look at the summary will notice that the estimate of the intercept (the average difference between the two pairs), the degrees of freedom and the t-value and the p-value are all the same as the value reported when using `t.test`.

In fact, all of the t-tests, and ANOVA (below) are kinds linear models and can be also fitted with `lm`.

Here is the paired t-test investigating gender differences in reaction time. You can see that the test statistics and coefficients match those obtained from `t.test`.

```
mod <- lm(Reaction ~ Gender, data = x)
anova(mod)
```

```
## Analysis of Variance Table
##
## Response: Reaction
##           Df  Sum Sq   Mean Sq F value  Pr(>F)
## Gender     1 0.01642 0.0164196  3.6482 0.06542 .
## Residuals 31 0.13952 0.0045008
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

```
summary(mod)
```

```
##
## Call:
## lm(formula = Reaction ~ Gender, data = x)
##
## Residuals:
##      Min       1Q   Median       3Q      Max
## -0.08938 -0.04558 -0.01258  0.03662  0.22542
```

```
##
## Coefficients:
##             Estimate Std. Error t value            Pr(>|t|)
## (Intercept)  0.34838    0.01581   22.03 <0.0000000000000002 ***
## GenderMale  -0.04480    0.02345   -1.91              0.0654 .
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## Residual standard error: 0.06709 on 31 degrees of freedom
## Multiple R-squared:  0.1053, Adjusted R-squared:  0.07643
## F-statistic: 3.648 on 1 and 31 DF,  p-value: 0.06542
```

14.8 Exercise: Sex differences in fine motor skills

Some people have suggested that there might be sex differences in fine motor skills in humans. Use the data collected on the class to address this topic using t-tests. The relevant data set is called `classData.csv`, and the columns of interest are `Gender` and `Precision`.

Carry out a two-sample t-test.

1) Plot the data (e.g. with a box plot, or histogram)
2) Formulate null and alternative hypotheses.
3) Use the `t.test` function to do the test.
4) Write a sentence or two describing the results.

14.9 Exercise: Therapy for anorexia

A study was carried out looking at the effect of cognitive behavioural therapy on weight of people with anorexia. Weight was measured in week 1 and again in week 8. Use a paired t-test to assess whether the treatment is effective.

The data is called `anorexiaCBT.csv`

The data are in "wide format". You may wish to convert it to "long format" depending on how you use the data. You can do that with the `pivot_longer` function, which rearranges the data:

```
anorexiaCBT_long <- anorexiaCBT %>%
  pivot_longer(
    cols = starts_with("Week"), names_to = "time",
    values_to = "weight"
  )
```

1) Plot the data (e.g. with an interaction plot like Figure 14.5)

2) Formulate a null and alternative hypothesis.

3) Use `t.test` to conduct a *paired* t-test.

4) Write a couple of sentences to report your result.

14.10 Exercise: Compare t-tests with randomisation tests (optional)

1. Try re-analysing some of the tests in this chapter as randomisation tests (or analyse the randomisation test data using `t.test`). Do they give the same results?
2. Try answering the question - "*are people who prefer dogs taller than those who prefer cats?*" using the `classData.csv`. Can you think of any problems with this analysis?

Chapter 15

Linear models with a single categorical explanatory variable

With the previous work on t-tests (and also with randomisation tests), you are now equipped to test for differences between two groups, or between one group and some fixed value. But what if there are more than two groups?

The answer is to use a one-way analysis of variance (ANOVA). Conceptually, this works the same way as a t-test.

15.1 One-way ANOVA

The one-way ANOVA is illustrated below with two cases (Figure 15.1). In both cases there are three groups. These could represent treatment groups in an experiment (e.g. different fertiliser addition to plants). In figure A, the three groups are very close, and the means are not significantly different from each other. In figure B, there is one group that stands apart from the others. The ANOVA will tell us whether *at least one* of the groups is different from the others.

After figuring out if at least one of the groups is significantly different from the others it is often enough to examine plots (or summary statistics) to see where the differences are (e.g. which group(s) are different from each other). In other cases it might be necessary to do follow up *post-hoc multiple comparison* tests. We will come to those later.

15.2 Fitting an ANOVA in R

New coffee machines use "pods" to make espresso. These have become much more popular than the traditional "bar machines". This data looks at the

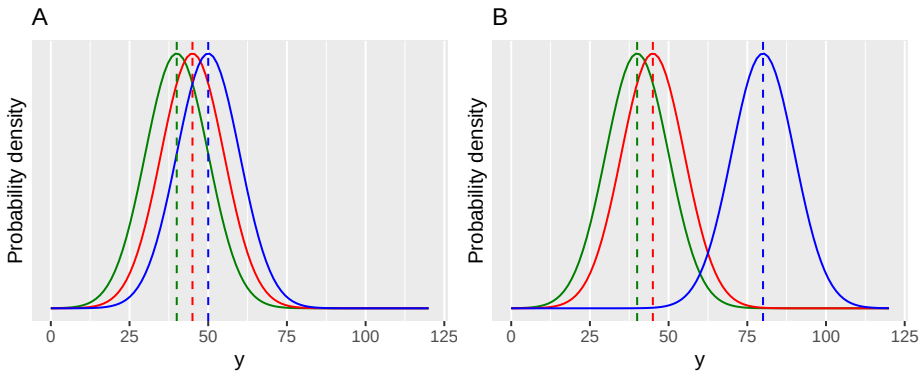


Figure 15.1: Visualising an ANOVA.

amount of "crema" or foam produced (a sign of quality!) using three methods: bar machines (BM), Hyper Espresso Pods (HIP) and Illy Espresso System (IES). Are any of these methods better than the others?

> Remember to load the `dplyr`, `magrittr` and `ggplot` packages, and to set your working directory correctly.

Import the data (`espresso.csv`) and look at it.

```
espresso <- read.csv("CourseData/espresso.csv",
                     stringsAsFactors = TRUE)
head(espresso)
```

```
##   foamIndx method
## 1    36.64     BM
## 2    39.65     BM
## 3    37.74     BM
## 4    35.96     BM
## 5    38.52     BM
## 6    21.02     BM
```

```
(ggplot(espresso, aes(x = method, y = foamIndx)) +
  geom_boxplot() +
  geom_jitter(width = 0.2))
```

You can see that the categorical explanatory variable ("method") defines the three treatment groups and has the three levels representing the different coffee types: BM, HIP and IES.

Let's first fit the ANOVA using R. One way ANOVAs are fitted using the `lm` function (`lm` stands for "linear model" - yes, an ANOVA is a type of linear model).

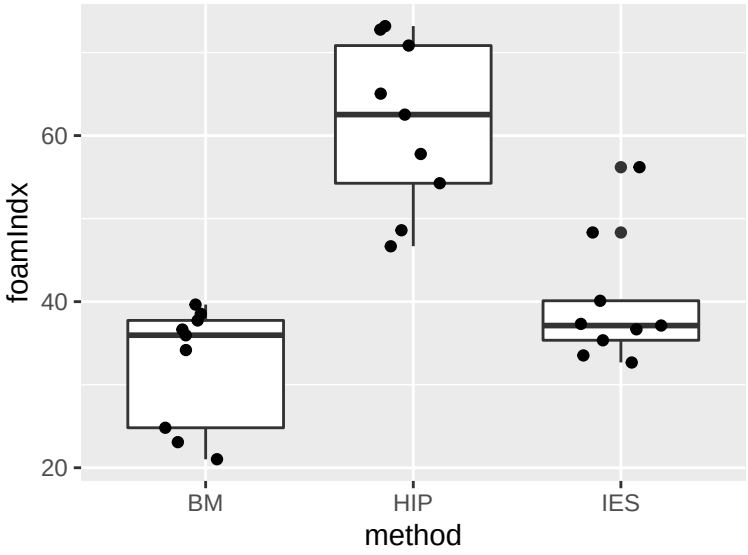


Figure 15.2: A box and whisker plot, with jittered points, for the espresso foam data.

```
foam_mod <- lm(foamIndx ~ method, data = espresso)
```

Before proceeding, we need to check the assumptions of the model. This can be done visually using the `autoplot` function in the `ggfortify` package. If you don't have the package installed, install it now (`install.packages("ggfortify")`).

```
library(ggfortify)
autoplot(foam_mod)
```

The main thing to look at here in Figure 15.3 is the "Q-Q" plot on the top right. We want those points to be approximately along the line. If that is the case, then it tells us that the model's residuals are normally distributed (this is one of the assumptions of ANOVA). We may cover these diagnostic plots more thoroughly later. You can find more details on pages 112-113 of the Beckerman et al textbook, or at the following if you are interested: https://data.library.virginia.edu/diagnostic-plots/.

Trust me, everything here looks great.

Now let's evaluate our ANOVA model. We do this using two functions: `anova` and `summary` (it sounds strange, but yes we do use a function called `anova` on our ANOVA model).

First, the `anova`. This gives us the following summary:

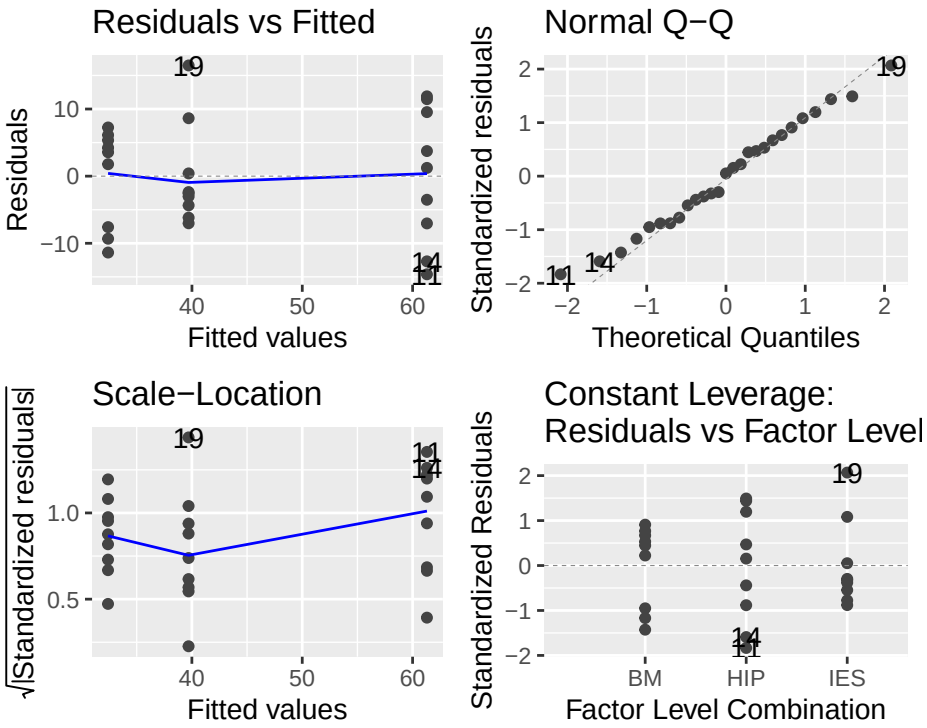


Figure 15.3: Diagnostic plots for the ANOVA model. This looks great.

```
anova(foam_mod)
```

```
## Analysis of Variance Table
##
## Response: foamIndx
##           Df Sum Sq Mean Sq F value       Pr(>F)
## method     2 4065.2 2032.59  28.413 0.0000004699 ***
## Residuals 24 1716.9   71.54
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

This gives some numbers (degrees of freedom, sum of squares, mean squares). These are the important values that go into calculating an *F value* (also called an F-statistic). We will not worry about these details now, except to say that large F-statistics mean that we are more certain that there is a difference between the groups (and that the p-value is smaller).

In this case, the F-value is 28.413.

As with the t-test, R compares this value to a theoretical distribution (a "table"), based on *two* degrees of freedom. The first one is number of groups minus one, i.e. 2.000 in this case. The second one is the overall sample size, minus the number of groups, i.e. 24.000, in this case.

This results in a p-value of 0.0000004699 (very highly significant!).

Based on this p-value we can reject the null hypothesis that there is no difference between groups. We might report this simple result like this:

The foam index varied significantly among groups (ANOVA: F = 28.413, d.f. = 2 and 24, p = 0.000).

15.2.1 Where are the differences?

This model output doesn't tell us *where* those differences are, nor does it tell us what the estimated mean values of foaminess are for the three groups: what are the effects? We need to dig further into the model to get to these details.

There are several ways to do this and we'll look at one of them.

We do this using the `summary` function.

```
summary(foam_mod)
```

```
##
## Call:
## lm(formula = foamIndx ~ method, data = espresso)
##
## Residuals:
##    Min     1Q Median     3Q    Max
## -14.62  -6.60   0.41   5.73  16.49
##
## Coefficients:
##             Estimate Std. Error t value        Pr(>|t|)
## (Intercept)   32.400      2.819  11.492 0.0000000000304 ***
## methodHIP     28.900      3.987   7.248 0.0000001728669 ***
## methodIES      7.300      3.987   1.831          0.0796 .
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## Residual standard error: 8.458 on 24 degrees of freedom
## Multiple R-squared:  0.7031, Adjusted R-squared:  0.6783
## F-statistic: 28.41 on 2 and 24 DF,  p-value: 0.0000004699
```

To properly interpret this output you need to understand something called "treatment contrasts". Essentially, contrasts define how model coefficients (the estimates made by the model) are presented in R outputs.

They are a bit hard to wrap your head aroudn and I STRONGLY recommend that you always do this with reference to a plot of the actual data, and the mean values for your groups. To do this you can use `group_by` and `summarise` to calculate the means for your the levels of your explanatory variable.

```
espresso %>%
  group_by(method) %>%
  summarise(gp_mean = mean(foamIndx))
```

```
## # A tibble: 3 x 2
##   method gp_mean
##   <fct>    <dbl>
## 1 BM        32.4
## 2 HIP       61.3
## 3 IES       39.7
```

Look at the coefficients of the model. Remember that you have three levels in your explanatory variable, but only two levels are shown in the summary. Which one is missing?

The "missing" group is the first one alphabetically (i.e. BM). The estimate (of the mean) for this group is labelled "`(Intercept)`" (with a value of 32.4. This is like a baseline or reference value, and the estimates for the other groups (HIP and IES), are *differences* between this baseline value and the estimated mean for those groups. In other words, the second group (HIP) is 28.9 more than 32.4 (32.4 + 28.9 = 61.3). Similarly, the third group (IES) is 7.3 more than 32.4 (32.4 + 7.3 = 39.7). Compare these values with the ones you got above using `summarise` - they should be the same.

This is illustrated below in Figure 15.3A. You can see that the coefficients of the model are the same as the lengths of the arrows that run from 0 (for the first level of method (BM), the Intercept) or *from* this reference value. It is often a good idea to sketch something like this on paper when you are trying to understand your model outputs!

Likewise, the t-values and p-values, are evaluating *differences between the focal group and this baseline*. Thus in this case, the comparisons (the "contrasts") are between the intercept (BM) and the second level (HIP), and the intercept (BM) and the third level (IES). There is no formal statistical comparison between HIP and IES.

You can see that it is very important to understand the levels of your explanatory variable, and how these relate to the summary outputs of the model. It can be useful to use the function `relevel` to manipulate the explanatory variable to make sure that the output gives you the comparisons you are interested in. Another simple trick would be to always ensure that your reference group (e.g. "control") comes first alphabetically and is therefore selected by R as the intercept (reference point).

For example, we can `relevel` the method variable so that the levels are re-ordered as HIP, BM, then IES so that the comparisons are between zero-HIP, HIP-BM and HIP-IES. (make sure that you understand this before proceeding).

```
# This is what the original data looks like:
levels(espresso$method)
```

```
## [1] "BM"  "HIP" "IES"
```

```
# releveling changes this by changing the reference.
espresso_2 <- espresso %>%
  mutate(method = relevel(method, ref = "HIP"))
levels(espresso_2$method)
```

```
## [1] "HIP" "BM"  "IES"
```

Now we can refit the model with this modified data set and see what difference that made:

```
foam_mod2 <- lm(foamIndx ~ method, data = espresso_2)
anova(foam_mod2)
```

```
## Analysis of Variance Table
##
## Response: foamIndx
##           Df Sum Sq Mean Sq F value       Pr(>F)
## method     2 4065.2 2032.59  28.413 0.0000004699 ***
## Residuals 24 1716.9   71.54
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

```
summary(foam_mod2)
```

```
##
## Call:
## lm(formula = foamIndx ~ method, data = espresso_2)
##
## Residuals:
##    Min     1Q Median     3Q    Max
## -14.62  -6.60   0.41   5.73  16.49
##
## Coefficients:
##             Estimate Std. Error t value             Pr(>|t|)
## (Intercept)   61.300      2.819  21.743 < 0.0000000000000002 ***
## methodBM     -28.900      3.987  -7.248          0.000000173 ***
## methodIES    -21.600      3.987  -5.417          0.000014501 ***
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## Residual standard error: 8.458 on 24 degrees of freedom
## Multiple R-squared:  0.7031, Adjusted R-squared:  0.6783
## F-statistic: 28.41 on 2 and 24 DF,  p-value: 0.0000004699
```

Now the coefficients in the model summary look different, but the model is actually the same. Compare the two graphs in Figure 15.4 - can you see the differences/similarities?

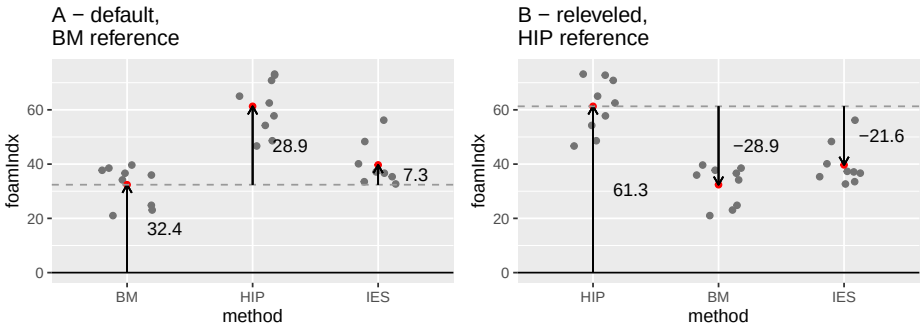


Figure 15.4: Comparison illustrating the difference between ANOVA models using (A) BM and (B) HIP as references in the espresso foam data set.

So, from the first of the model outputs above you can say that BM is significantly different than HIP (t= 7.248, p $<$ 0.0001), but that BM is not significantly different from IES (t = 1.831, p = 0.0796). Then, from the second one you can see that HIP is significantly different from IES (t=-5.417, p $<$ 0.0001). You could write this into the main text, include the information in a figure caption (i.e. add it to Figure 15.2.

e.g. *The foam index varied significantly among groups (ANOVA: F = 28.413, d.f. = 2 and 24, p = 0.000). The pairwise comparisons in the ANOVA model showed that means of BM and HIP were significantly different (t= 7.248, p $<$ 0.0001), as were those of HIP and IES (t=-5.417, p $<$ 0.0001), but the BM-IES comparison showed no significant difference (t= 1.831, p= 0.0796).*

15.2.2 Tukey's Honestly Significant Difference (HSD)

An alternative way to approach these comparisons between groups is to use something called a **post-hoc multiple comparison test**. The words "post-hoc" mean "after the event" – i.e. after the ANOVA in this case – while the "multiple comparison" refers to the (potentially many) pairwise comparisons that you would like to make with an ANOVA. One of the most widely used post-hoc tests is called Tukey's Honestly Significant Difference (Tukey HSD) test. There is a convenient R package called `agricolae` that will do these for you.

```
# You only need to do this once!
install.packages("agricolae")
```

When you have the package installed, you can load it (using `library`). Then you can run the Tukey HSD test using the function `HSD.test`. The first argument for the function is the name of the model, followed by the name of the variable

you are comparing (in this case `method`) and finally `console = TRUE` tells the function to print the output to your computer screen.

```
library(agricolae)
HSD.test(foam_mod2, "method", console = TRUE)
```

```
##
## Study: foam_mod2 ~ "method"
##
## HSD Test for foamIndx
##
## Mean Square Error:  71.5383
##
## method,  means
##
##     foamIndx       std r   Min   Max
## BM      32.4  7.300060 9 21.02 39.65
## HIP     61.3 10.100604 9 46.68 73.19
## IES     39.7  7.700768 9 32.68 56.19
##
## Alpha: 0.05 ; DF Error: 24
## Critical Value of Studentized Range: 3.531697
##
## Minimun Significant Difference: 9.957069
##
## Treatments with the same letter are not significantly different.
##
##     foamIndx groups
## HIP     61.3      a
## IES     39.7      b
## BM      32.4      b
```

The output is long-winded, and the main thing to look at is the part at the end. The key to understanding this is actually written in the output, "`Treatments with the same letter are not significantly different`".

You could include these in a figure or a table with text like, *"Means followed by the same letter did not differ significantly (Tukey HSD test, p>0.05)"*.

15.3 ANOVA calculation "by hand".

> The following is an optional exercise designed to help you understand how ANOVA works.

The ANOVA calculation involves calculating something called an F-value or F-statistic (the F stands for Fisher, who invented ANOVA). This is similar to the

t-statistic in that it is a ratio between two quantities, in this case variances. In ANOVA, the F-statistic is calculated as the **treatment variance** divided by the **error variance**.

What does that mean? Let's consider the espresso data set again.

In the Figure 15.4, you can see on the left (A) the black horizontal line which is the overall mean foam index. The vertical lines are the "errors" or departures from the overall mean value, colour coded by treatment (i.e. method). These can be quantified by summing up their square values (squaring ensures that the summed values are all positive). We call this quantity the *Total Sum of Squares (SSTotal)*. If there is a lot of variation, the sum of squares will be large, if there is little variation the sum of squares will be small.

On the right hand side (Figure 15.4B) we see the same data points. However, this time the horizontal lines represent the treatment-specific mean values, and the vertical lines illustrate the errors from these mean values. Again we can sum up these as sum of squares, which we call the *Error Sum of Squares (SSError)*.

The difference between those values is called the *Treatment Sum of Squares (SSTreatment)* and is the key to ANOVA - it represents the importance of the treatment:

$$SSTreatment = SSTotal - SSError$$

.

If that doesn't make sense yet, picture the case where the treatment-specific means are all very similar, and are therefore very close to the overall mean. Now the difference between the *Total Sum of Squares* and the *Error Sum of Squares* will be small. Sketch out a couple of examples with pen on paper if that helps. You should now see that you can investigate differences among means by looking at variation.

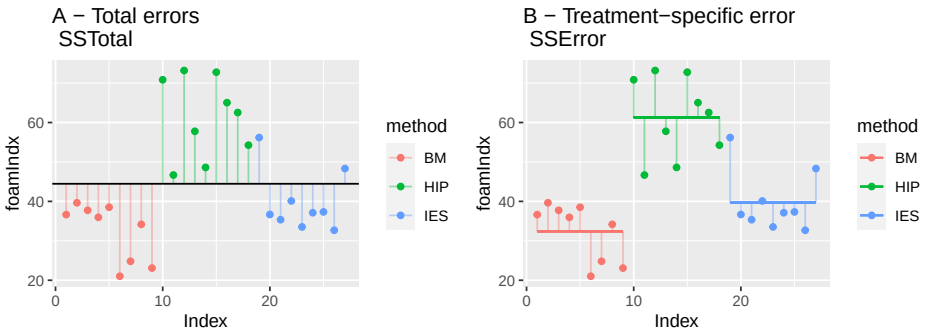


Figure 15.5: The relative size of the squared residual errors from the overall mean (SSTotal) (A) and from the treatment-specific means (SSError) (B) tell us about the importance of the treatment variable. The difference between the two values is the "treatment sum of squares".

In the following I will show how these calculations can be done "by hand" in R. The purpose of showing you this is to demonstrate exactly how the `lm` model that you fitted above works, and prove to yourself that it is not rocket science...

you will never need to do this in real life, because you have the wonderful `lm` function.

Here goes...

First, calculate the total sum of squares:

```
(SSTotal <- sum((overallMean - espresso$foamIndx)^2))
```

```
## [1] 5782.099
```

Now calculate the group-specific means:

```
(groupMeans <- espresso %>%
  group_by(method) %>%
  summarise(groupMean = mean(foamIndx)) %>%
  pull(groupMean))
```

```
## [1] 32.4 61.3 39.7
```

Now add those group-specific mean values to the dataset using `left_join` so that you can calculate the group-specific errors.

```
espresso <- left_join(espresso, espresso %>%
  group_by(method) %>%
  summarise(groupMean = mean(foamIndx))) %>%
  mutate(groupSS = (foamIndx - groupMean)^2)

head(espresso)
```

```
##   foamIndx method groupMean  groupSS
## 1    36.64     BM      32.4  17.9776
## 2    39.65     BM      32.4  52.5625
## 3    37.74     BM      32.4  28.5156
## 4    35.96     BM      32.4  12.6736
## 5    38.52     BM      32.4  37.4544
## 6    21.02     BM      32.4 129.5044
```

Then, to calculate the errors:

```
(SSError <- espresso %>%
  summarise(sum(groupSS)) %>%
  pull())
```

```
## [1] 1716.919
```

From there, you can calculate the **Treatment Sum of Squares**

```
(SSTreatment <- SSTotal - SSError)
```

```
## [1] 4065.18
```

So far, so good - but we can't just look at the ratio of SSTreatment/SSError, because sum of square errors always increase with sample size. We can account for this by dividing

We need to take account of sample size (degrees of freedom) by dividing these sum of squares by the degrees of freedom to give us variances. There are 3 treatment groups and 9 samples per group. Therefore there are 2 degrees of freedom for the treatment, and 8 degrees of freedom per each of the three treatments, giving a total of 8*3 = 24 error degrees of freedom.

Now we need to correct for degrees of freedom, which will give us variances.

```
(meanSSTreatment <- SSTreatment / 2)
```

```
## [1] 2032.59
```

```
(meanSSError <- SSError / 24)
```

```
## [1] 71.5383
```

The F-statistic is then the ratio of these values.

```
(Fstat <- meanSSTreatment / meanSSError)
```

```
## [1] 28.41261
```

We can "look up" the p-value associated with this F-statistic using the `pf` function (`pf` stands for probability of f) like this:

```
1 - pf(Fstat, df1 = 2, df2 = 24)
```

```
## [1] 0.0000004698636
```

As you can see, the method is a bit laborious and time consuming but it is conceptually fairly straightforward - it all hinges on the ratio of variation due to treatment effect vs. overall variation. Signal and noise.

15.4 Exercise: Apple tree crop yield

An experiment was conducted to look at the effect of tree spacing on apple crop yield (total kg per tree of apples between 1975-1979) in different spacing conditions (i.e. distances between trees). There were 40 trees in each treatment group. The spacing was 6, 10 and 14 feet, and should be treated as a categorical variable. There may be some `NA` missing yield values.

Import the data (`apples.csv`) and analyse it using the techniques you have learned in the ANOVA lecture, and the previous chapter, to answer the question "What is the effect of tree spacing on apple yields?"

1) Import and look at the data (e.g. `summary` or `str` or `head`)
2) Ensure that the explanatory variable (`spacing`) is defined as a categorical variable (i.e. a "factor", in R-speak). You can use `mutate` and `as.factor` functions for this.
3) Plot the data using `ggplot` (a box plot with (optionally) added jittered points would be good).
4) Fit an ANOVA model using `lm`.
5) Check the model using a diagnostic plot (i.e. using `autoplot` from the `ggfortify` package).
6) Use `anova` to get the ANOVA summary.
7) You should see that there are differences among treatments. But where are those differences? Use `summary` on your model to find out.
8) Use a Tukey test to make all the pairwise comparisons among the treatments.
9) Write a few sentences that summarise your findings. What biological processes do you think drive the effects that you have detected?
10) Optional. Instead of using a Tukey test, use the alternative "relevel" approach to make the missing comparison.

If you get this far, try using the ANOVA approach on one of the previous t-test examples (remember that ANOVA can be used when your single explanatory variable has TWO or more levels). You should notice that the results are the same whether you use the `t.test` function or the ANOVA approach with `lm`.

Chapter 16

Linear models with a single continuous explanatory variable

Linear regression models, at their simplest, are a method of estimating the linear (straight line) relationships between two continuous variables. As an example, picture the relationship between two variables height and hand width (Figure 16.1). In this figure there is a clear relationship between the two variables, and the straight line running through the cloud of data points is the fitted linear regression model.

The aim of linear regression is to (1) determine if there is a meaningful statistical relationship between the explanatory variable(s) and the response variable, and (2) to quantify those relationships by estimating the characteristics of those relationships. These characteristics include the slope and intercepts of fitted models, and the amount of variation explained by variables in the model.

16.1 Some theory

To understand linear regression models it is important to know that the equation of a straight line is $y = ax + b$. In this equation, y is the response variable and x is the explanatory variable, and a and b are the slope and intercept of the line with the vertical axis (y-axis). These (a and b) are called **coefficients**. These are illustrated in Figure 16.2.

When looking at data points on a graph, unless all of the data points are arranged perfectly along a straight line, there will be some distance between the points and the line. These distances, measured parallel to the vertical axis, are called residuals (you have encountered them before in this course). These residuals represent the variation left after fitting the line (a linear model) through the data. Because we want to fit a model that explains as much variation as possible, it is intuitive that we should wish to minimise this residual variation.

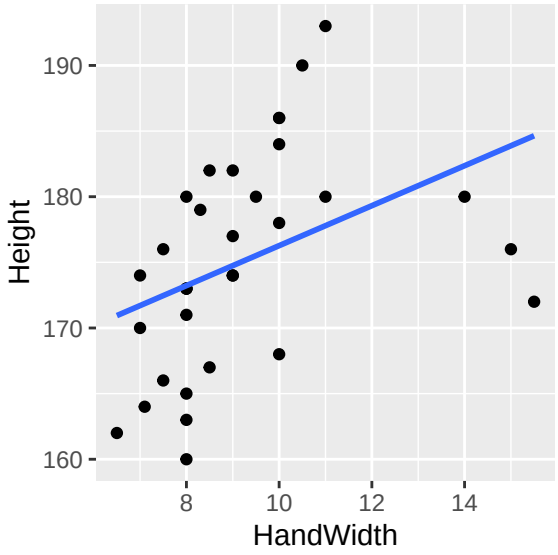


Figure 16.1: A linear regression model fitted through data points.

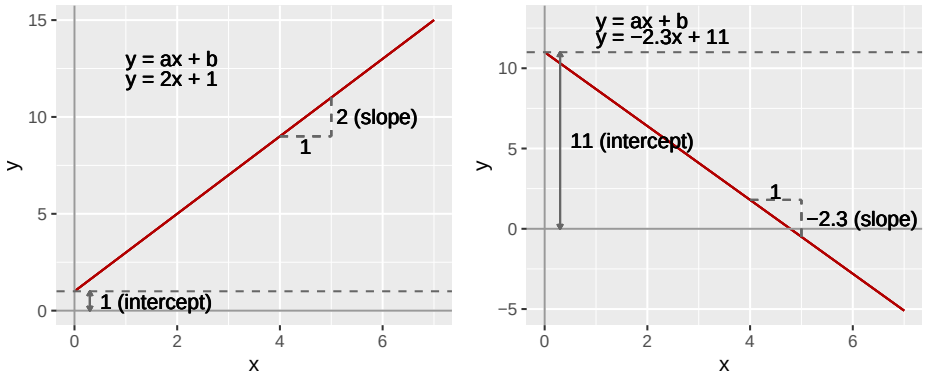


Figure 16.2: The equation of straight lines.

One way of doing this is by minimising the sum of squares of the residuals (again, you have come across this concept a few times before). In other words, we add up the squares of each of the residuals. We square the values, rather than simply adding up the residuals themselves because we want to ensure that the positive and negative values don't cancel each other out (a square of a negative number is positive). This method is called **least squares** regression and is illustrated in Figure 16.3: Which is the best fitting line?

In fact, these residuals represent "error" caused by factors including measurement error, random variation, variation caused by unmeasured factors etc. This error term is given the label, ϵ. Thus we can write the model equation as:

$$y = ax + b + \epsilon$$

Sometimes, this equation is written with using the beta symbol (β) for the

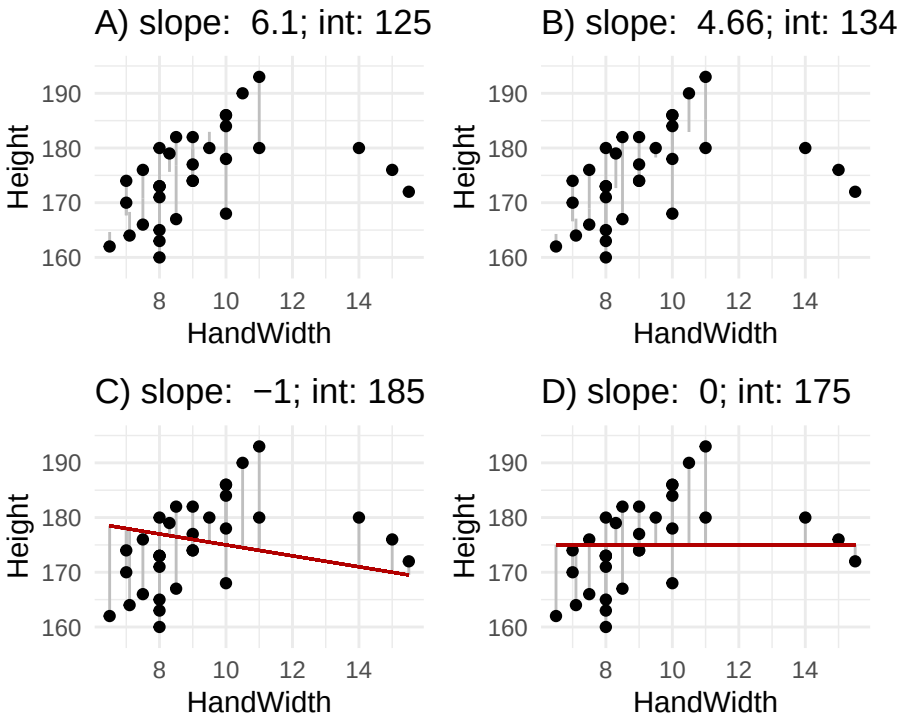


Figure 16.3: Residuals and least squares: which is the best fitting line?

coefficients, so that the slope is β_0 and the intercept is β_1 for example.

$$y = \beta_0 x + \beta_1 + \epsilon$$

The idea is that this equation, and its coefficients and error estimates, describe the relationship we are interested in (including the error or uncertainty).

Together this information allows us to not only determine if there **is** a statistically significant relationship, but also what the nature of the relationship is, and the uncertainty in our estimates.

16.2 Evaluating a hypothesis with a linear regression model

Usually, the most important hypothesis test involved with a linear regression model relates to the slope: **is the slope coefficient significantly different from 0?**, or should we accept the null hypothesis that the slope is no different from 0.

Sometimes hypotheses like this are a bit boring, because we already know the answer before collecting and analysing the data. What we usually **don't** know is the nature of the relationship (the slope, intercept, their errors, and amount

of variation explained). Usually it is more interesting and meaningful to focus on those details.

The following example, where we focus on the relationship between hand width and height, is one of these "boring" cases: we already know there is a relationship. Nevertheless, we'll use this example because it helps us understand how this hypothesis test works.

The aim of this section is to give you some intuition on how the hypothesis test works.

We can address the slope hypothesis by calculating an F-value in a similar way to how we used them in ANOVA. Recall that F-values are ratios of variances. To understand how these work in the context of a linear regression we need to think clearly about the slope hypothesis: The **null hypothesis** is that the slope is **not** significantly different to 0 (that the data can be explained by random variation). The **alternative hypothesis** is that the slope is significantly different from 0.

The first step in evaluating these hypotheses is to calculate what the **total sum of squares**[1] is when the null hypothesis is true (Figure 16.4A) - this value is the total variation that the model is trying to explain.

Then we fit our model using least squares and figure out what the **residual sum of squares** is from this model (Figure 16.4B). This is the amount of variation left after the model has explained *some* of the total variation - it is sometimes called *residual error*, or simply *error*.

The difference between these two values is the **explained sum of squares**, which measures the amount of variation in y explained by variation in x. The rationale for this is that the model is trying to explain total variation. After fitting the model there will always be some unexplained variation ("*residual error*") left. If we can estimate total variation and unexplained variation, then the amount of variation explained can be calculated with a simple subtraction:

$$Total = Explained + Residual$$

… and, therefore …

$$Explained = Total - Residual$$

Before using these values we need to standardise them to control for sample size. This is necessary because sum of squares will *always* increase with sample size. We make this correction by dividing our sum of squares measures by the degrees of freedom. The d.f. for the **explained sum of squares** is 1, and the d.f. for the **residual sum of squares** is the number of observations minus 2. The result of these calculations is the **mean explained sum of squares** (mESS) and the **mean residual sum of squares** (mRSS). These "mean" quantities are **variances**, and the ratio between them gives us the **F-value**. Notice that this is very similar to the variance ratio used in the ANOVA.

$$F = \frac{mESS}{mRSS}$$

[1] sum of squares is simply a way to estimate variation.

If the *explained variance* (mESS) is large compared to the *residual error variance* (mRSS), then F will be large. The size of F tells us how likely or unlikely it is that the null hypothesis is true. When F is large, the probability that the slope is significantly different from 0 is high. To obtain the actual probabilities, the F-value must be compared to a theoretical distribution which depends on the two degrees of freedom (explained and residual d.f.). Once upon a time you would have looked this up in a printed table, but now R makes this very straightforward.

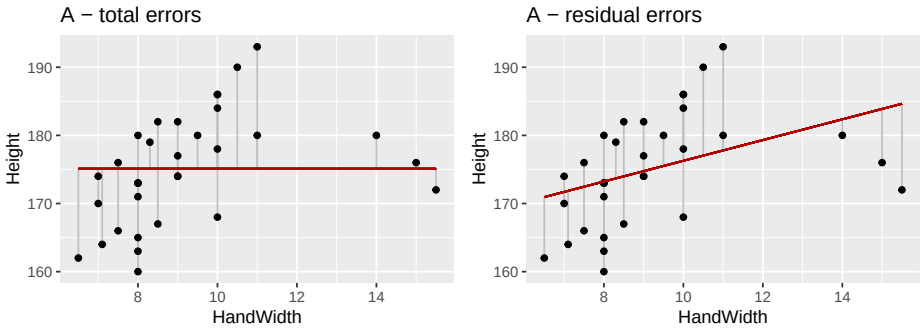


Figure 16.4: (A) the total variation around the overall mean Height value (B) the residual error of the model.

16.3 Assumptions

These models have similar assumptions to the other linear models[2]. These are (1) that the relationship between the variables is linear (hence the name); (2) that the data are continuous variables; (3) that the observations are randomly sampled; (4) that the errors in the model (the "residuals") can be described by a normal distribution; and (5) and that the errors are "homoscedastic" (that they are constant through the range of the data). You can evaluate these things by looking at diagnostic plots after you have fitted the model. See page 112-113 in GSWR for a nice explanation.

16.4 Worked example: height-hand width relationship

Let's now use R to fit a linear regression model to estimate the relationship between hand width and height. One application for such a model could be to predict height from a hand print, for example left at a crime scene. I am restricting my analysis to 2019 data, but you could do it for any year (or all years, but you might need to first get rid of some outliers using `filter`).

[2] t-tests, ANOVA and linear regression are all types of linear model, mathematically

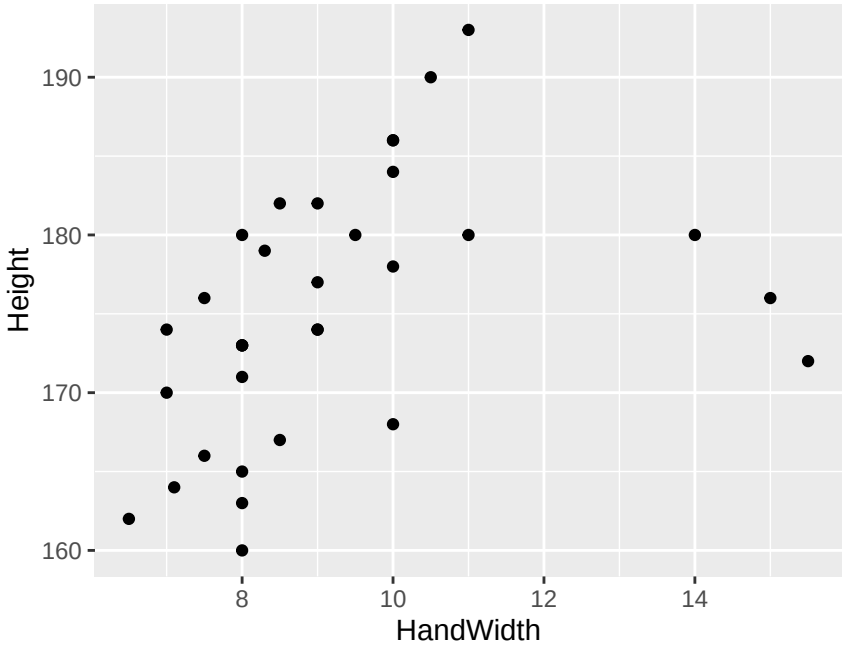
Remember to load the `dplyr`, `magrittr` and `ggplot` packages, and to set your working directory correctly.

First, load the data:

```
classData <- read.csv("CourseData/classData.csv") %>%
  filter(Year == 2019)
```

We should then plot the data to make sure it looks OK.

```
ggplot(classData, aes(x = HandWidth, y = Height)) +
  geom_point()
```



This looks OK, and the relationship looks fairly linear. Now we can fit a model using the `lm` function (same as for ANOVA!).[3]

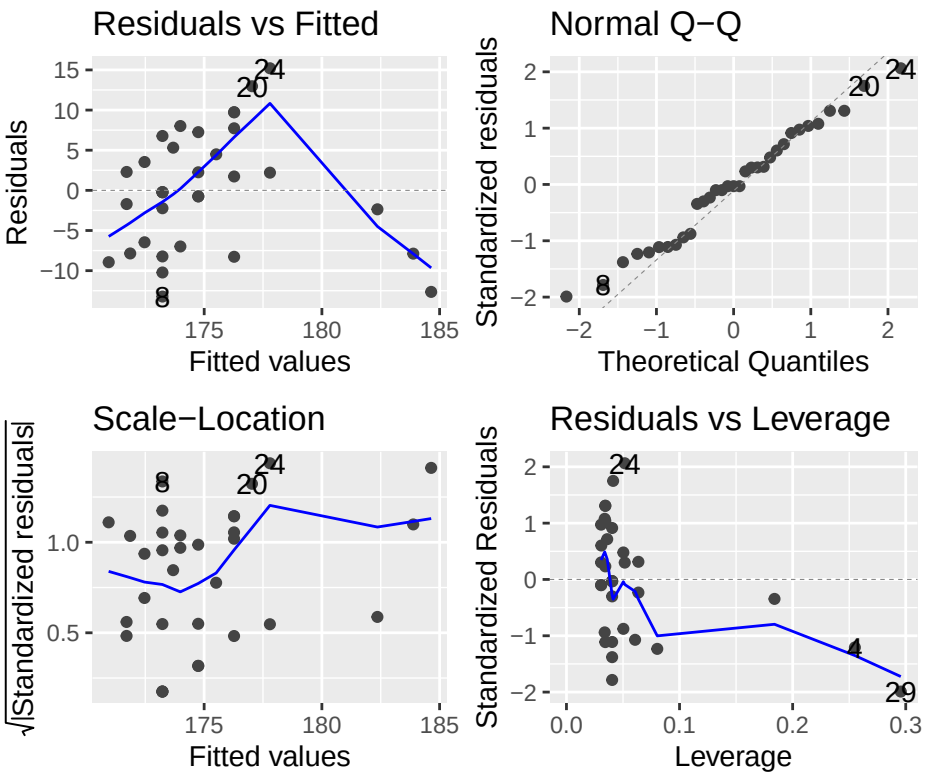
The response variable is always the one we would like to predict, in this case `Height`. The explanatory variable (sometimes called the predictor) is `HandWidth`. These are added to the model using a formula where they are separated with the `~` ("tilde") symbol: `Height ~ HandWidth`. In the model expression, we also need to tell R where the data are using the `data =` argument. We can save the model as an R object by naming it e.g. `mod_A <-`.

[3] R knows that this is a linear regression rather than an ANOVA because the explanatory variable is numeric rather than categorical - smart!.

```
mod_A <- lm(Height ~ HandWidth, data = classData)
```

Before proceeding further we should evaluate the model using a diagnostic plot. We can do this using the `autoplot` function in the `ggfortify` package (you may need to install and/or load the package).

```
library(ggfortify)
autoplot(mod_A)
```



These diagnostic plots allow you to check that the assumptions of the model are not violated. On the left are two plots which (more or less) show the same thing. They show how the residuals (the errors in the model) vary with the predicted value (height). Looking at the plots allows a visual test for constant variance (homoscedasticity) along the range of the data. In an ideal case, there should be no pattern (e.g. humps) in these points. On the top right is the QQ-plot which shows how well the residuals match up to a theoretical normal distribution. In an ideal case, these points should line up on the diagonal line running across the plot. The bottom right plot shows "leverage" which is a measure of how much influence individual data points have on the model. Outliers will have large leverage and can mess up your model. Ideally, the points here should be in a cloud, with no points standing out from the others. Please read the pages

112-113 in the textbook GSWR for more on these. In this case, the model looks pretty good.

Now that we are satisfied that the model doesn't violate the assumptions we can dig into the model to see what it is telling us.

To test the (slightly boring) slope hypothesis we use the `anova` function (again, this is the same as with the ANOVA).

```
anova(mod_A)
```

```
## Analysis of Variance Table
##
## Response: Height
##           Df  Sum Sq Mean Sq F value Pr(>F)
## HandWidth  1  343.86  343.86  6.0059 0.0201 *
## Residuals 31 1774.86   57.25
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

When you run this function, you get a summary table that looks exactly like the one you got with an ANOVA. There are degrees of freedom (`Df`), Sums of Squares (`Sum Sq`), Mean Sums of Squares (`Mean Sq`) and the `F value` and p-value (`Pr(>F)`). The most important parts of this table are the F value (6.0059) and the p-value (0.02009856): as described above, large F values lead to small p-values. This tells us that it is VERY unlikely that the null hypothesis is true and we should accept the alternative hypothesis (that height is associated with hand width)

We could report the results of this hypothesis test like this: *There was a statistically significant association between hand width and height ($F = 6.0059$, d.f. = 1,31, $p < 0.001$)*

Now we can dig deeper by asking for a `summary` of the model.

```
summary(mod_A)
```

```
##
## Call:
## lm(formula = Height ~ HandWidth, data = classData)
##
## Residuals:
##      Min       1Q   Median       3Q      Max
## -13.2276  -6.9886  -0.2276   5.3158  15.2064
##
## Coefficients:
##             Estimate Std. Error t value            Pr(>|t|)
## (Intercept)  161.052      5.878  27.398 <0.0000000000000002 ***
## HandWidth      1.522      0.621   2.451              0.0201 *
```

```
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## Residual standard error: 7.567 on 31 degrees of freedom
## Multiple R-squared:  0.1623, Adjusted R-squared:  0.1353
## F-statistic: 6.006 on 1 and 31 DF,  p-value: 0.0201
```

This summary has a lot of information. First we see `Call` which reminds us what the formula we have used to fit the model. Then there is some summary information about the residuals. Ideally these should be fairly balanced around 0 (i.e. the `Min` value should be negative but with the same magnitude as `Max`). If they are **wildly** different, then you might want to check the data or model. In this case they look OK.

Then we get to the important part of the table - the `Coefficients`. This lists the coefficients of the model and shows first the `Intercept` and then the slope, which is given by the name of the explanatory variable (`HandWidth` here). For each coefficient we get the `Estimate` of its value, and the uncertainty in that estimate (the standard error ('Std. Error)).

These estimates and errors are each followed by a `t value` and a p-value (`Pr(>|t|)`). These values provide a test of whether the slope/intercept is different from zero. In this case they both are. The t-tests are indeed doing t-tests of these estimates, in the same way that a regular t-test works, so that the significance depends on the ratio between signal (the estimate) and the noise (the standard error). This is illustrated for the coefficient estimates for our model in Figure 16.5.

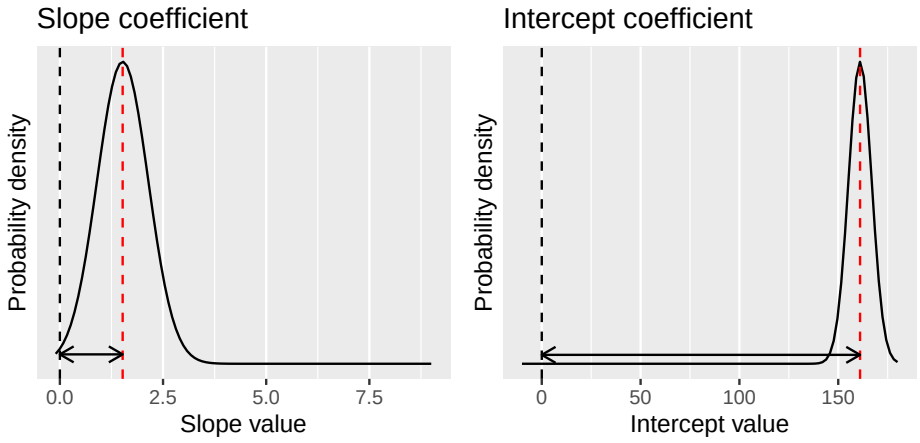


Figure 16.5: Illustration of the coefficient estimates for our model. The peak of the distribution is at the coefficient estimate, and the spread of the distribution indicates the standard error of the mean for the estimate. The statistical significance of the coefficient is determined by the degree of overlap with 0.

The summary then gives some information about the amount of residual variation left after the model has been fitted (this is the ϵ term in the equations at the start of this chapter). Then we are told what the R^2 value is 0.1623.

The adjusted R^2 is for use in multiple regression models, where there are many explanatory variables and should not be used for this simple regression model. So what does R^2 actually mean?

R^2 is the square of the correlation coefficient r and is a measure of the amount of variation in the response variable (Height) that is explained by the model. If all the points were sitting on the regression line, the R^2 value would be 1. This idea is illustrated in Figure 16.6.

We could describe the model like this:

There is a statistically significant association between hand width and height ($F = 6.0059$, $d.f. = 1,31$, $p < 0.001$) The equation of the fitted model is: Height = 1.52(± 0.62) × HandWidth + 161.05(± 5.88). The model explains 16% of the variation in height ($R^2 = 0.162$).

... or maybe, *The model, which explained 16% of the variation in height, showed that the slope of the relationship between hand width and height is 1.52 ± 0.62 which is significantly greater than 0 ($t = 27.40$, $p < 0.01$)*

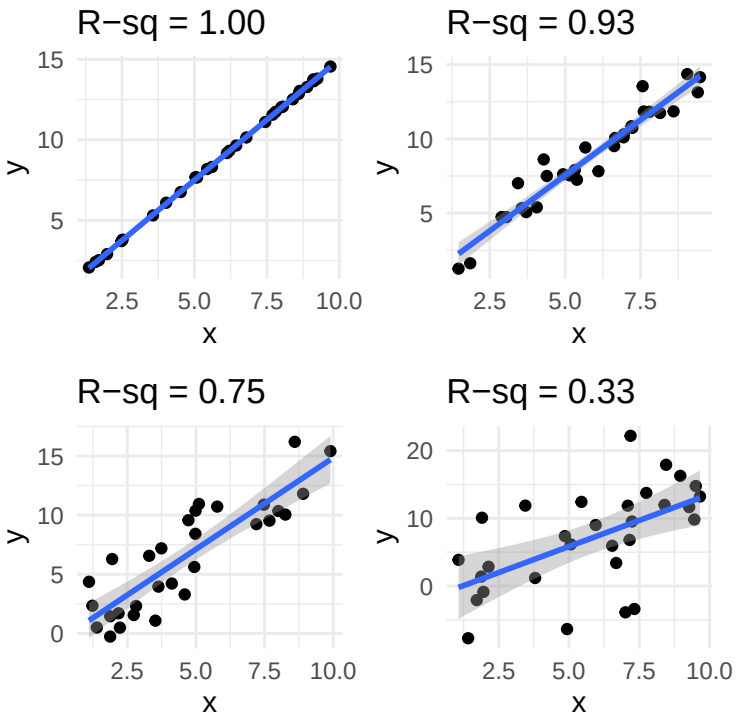
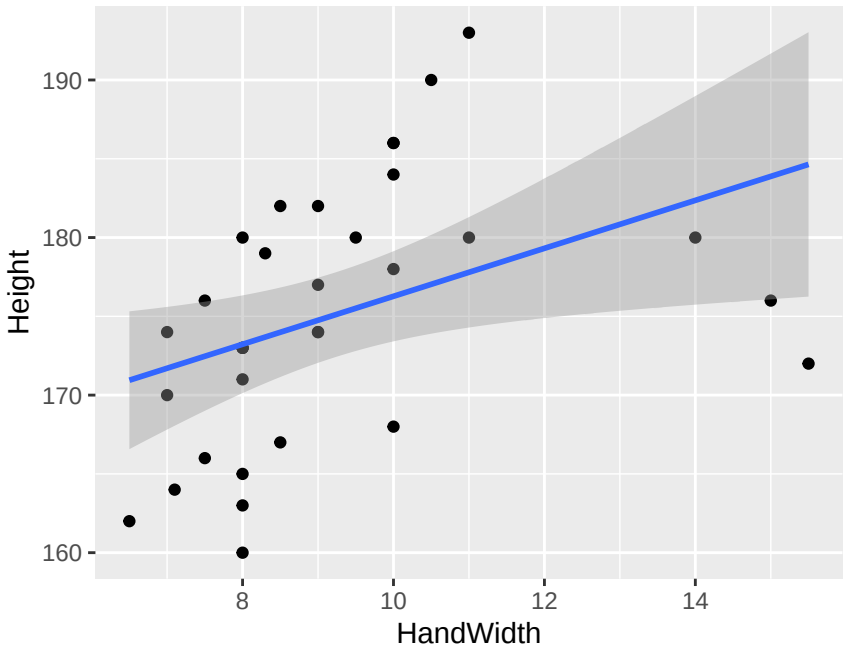


Figure 16.6: An illustration of different R-squared values.

A plot is usually a good idea because it is easier for the reader to interpret than an equation, or coefficients. The `ggplot2` package has a neat and simple function called `geom_smooth` which will add the fitted regression line to simple models like this. For linear regression models you simply need to tell it to use `method = "lm"`. This will plot the fitted regression model, and will add, by default" a shaded "ribbon" which represents the so called "95% confidence interval" for the fitted values. These are 2 time the standard error.

```
ggplot(classData, aes(x = HandWidth, y = Height)) +
  geom_point() +
  geom_smooth(method = "lm")
```



Question: If police find the 9.8cm wide hand print at a crime scene, what is your best guess of the height of the person involved?

16.5 Exercise: Chirping crickets

Male crickets produce a "chirping" sound by rubbing the edges of their wings together: the male cricket rubs a sharp ridge on his wing against a series ridges on the other wing. In a 1948 study on striped ground cricket (*Allonemobius fasciatus*), the biologist George W. Pierce recorded the frequency of chirps (vibrations per second) in different temperature conditions.

Crickets are ectotherms so their physiology and metabolism is influenced by temperature. We therefore believe that temperature might have an effect on their chirp frequency.

The data file `chirps.csv` contains data from Pierce's experiments. Your task is to analyse the data and find (1) whether there is a statistically significant relationship between temperature and chirp frequency and (2) what that relationship is.

The data has two columns - `chirps` (the frequency in Hertz) and `temperature` (the temperature in Fahrenheit). You should express the relationship in Celsius.

1) Import the data
2) Use `mutate` to convert Fahrenheit to Celsius (Google it)
3) Plot the data
4) Fit a linear regression model with `lm`
5) Look at diagnostic plots to evaluate the model
6) Use `anova` to figure out if the effect of temperature is statistically significant.
7) Use `summary` to obtain information about the coefficients and R^2-value.
8) Summarise the model in words.
9) Add model fit line to the plot.
10) Can I use cricket chirp frequency as a kind of thermometer?

Chapter 17

Linear models with categorical and continuous explanatory variables

In the previous chapter we looked at linear models where there is a continuous response variable and two categorical explanatory variables (we call this type of linear model two-way ANOVA). In this chapter we will look at linear models where the explanatory variables are both continuous and categorical. You can think of these as a kind of cross between ANOVA and linear regression. These type of models are often given the name "*ANCOVA*" or *Analysis of Covariance*.

In a simple case, you might be interested in a model with a continuous response variable (e.g. height) and continuous and a categorical explanatory variables (e.g. hand width and gender). The categorical variable may have any number of levels, but the simplest case is with two (e.g. gender with male and female levels).

Some of these different possible outcomes of this type of analysis are illustrated in Figure 18.1. We might see that neither of the two explanatory variables has a significant effect. We might see that one of them does but not the other one. We might see an interaction effect (where the effect of one variable (e.g. hand width) depends on the other (e.g gender). We might also see an interaction effect but no main effect.

17.1 The height ~ hand width example.

In a previous class (linear regression) you explored the relationship between hand width and height. The aim there was (1) to determine if the relationship (i.e. the slope) was significantly different from 0. and (2) to make an estimate of what the equation of the relationship would be so you could make predictions of height from hand width.

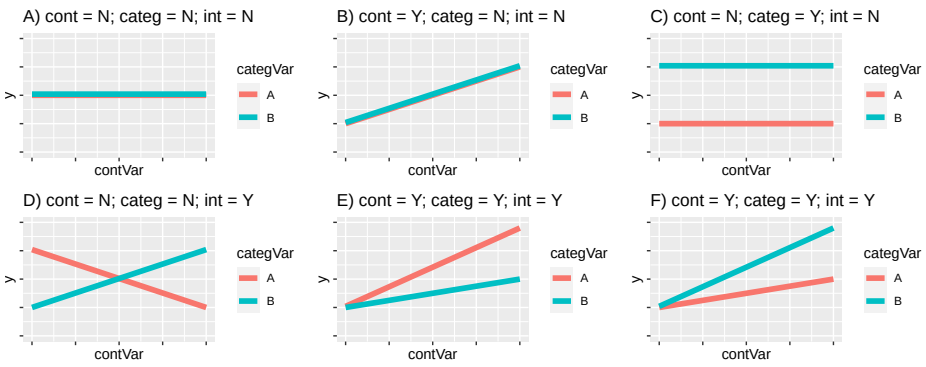


Figure 17.1: Some potential results of the experiment. There may be a significant effect (or not) of both of the main effects (diet and genotype) and there may be a significant interaction effect (or not).

Here we will extend that example by asking whether there are differences between males and females. I am restricting my analysis to 2019 data, but you could do it for any year (or all years, but you might need to first get rid of some outliers using `filter`).

> Remember to load the `dplyr`, `magrittr` and `ggplot` packages, and to set your working directory correctly.

We'll begin by plotting the data (Figure 17.2).

```
classData <- read.csv("CourseData/classData.csv") %>%
  filter(Year == 2019)

(A <- ggplot(classData, aes(
  x = HandWidth, y = Height,
  colour = Gender
)) +
  geom_point() +
  geom_smooth(method = "lm", se = FALSE))
```

```
# This shows the ANCOVA model
# before we have even fit it!
```

You can see that our two continuous variables, `Height` (the response variable) and `HandWidth` (one of the explanatory variables) are associated: There is an overall positive relationship between `HandWidth` and `Height` You can also see that `Gender` (the categorical explanatory variable) is important: males tend to be taller than females for any given hand width. For example, a female with hand width of 9cm is ~172cm tall while a male would be about 180cm tall. This

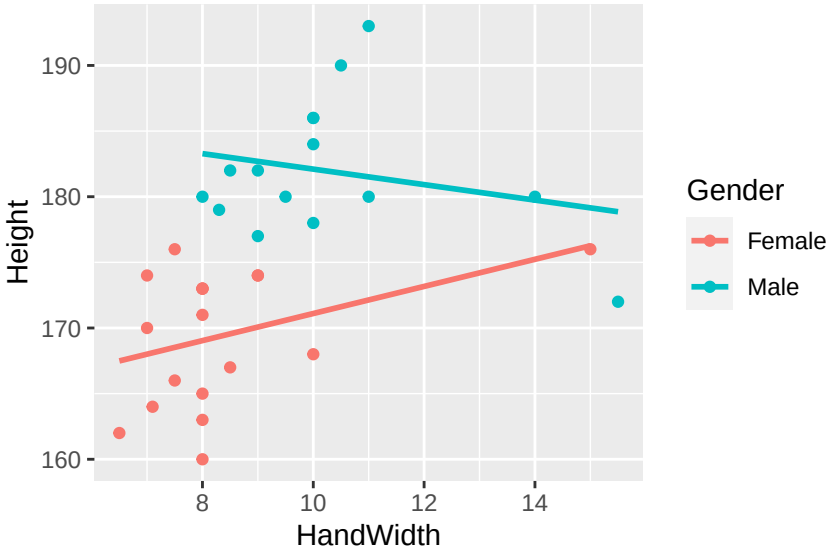


Figure 17.2: ANCOVA on hand width vs. height data in males and females

shows us that males have a higher **intercept** than females. There is also a **slight** difference in the slope of the relationship, with males having a slightly steeper slope than females. We already know that the overall relationship between hand width and height is significant (from the linear regression chapter). These new observations leave us with the following additional questions: (1) are the intercepts for males and females significantly different? (2) are the slopes for males and females significantly different (or would a model with a single slope, but different intercepts be better)?

Now we can fit our model using the `lm` function. The model formula is `Height ~ HandWidth + Gender + HandWidth:Gender`. The `HandWidth` and `Gender` are the so called **main effects** while `HandWidth:Gender` represents the interaction between them (i.e. it is used to address the question "*does the effect of hand width differ between the sexes?*"). R knows that is fitting an ANCOVA type model rather than a two-way ANOVA because it knows the type of variables that it is dealing with. You can see this if you ask R to tell you what the `class` of the variables are:

```
class(classData$Gender)
```

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

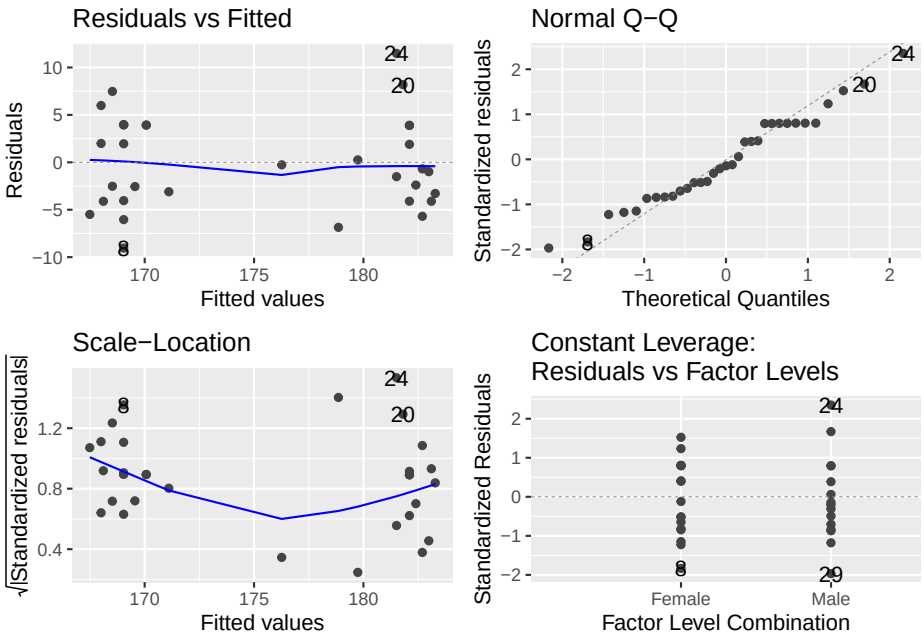
```
class(classData$HandWidth)
```

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

```
mod_A <- lm(Height ~ HandWidth + Gender + HandWidth:Gender,
  data = classData
)
```

The first step should, as before, be to check out the diagnostic plots. We should not read to much into these in this case, because we have a small sample size. Nevertheless, lets keep with good habits:

```
library(ggfortify)
autoplot(mod_A)
```



These look good. No evidence of non-normality in the residuals, no heteroscedasticity and no weird outliers.

17.2 Summarising with `anova`

Now we can get the `anova` table of our ANCOVA model (yes, I know that sounds strange).

```
anova(mod_A)
```

```
## Analysis of Variance Table
##
## Response: Height
```

```
##                   Df Sum Sq Mean Sq F value      Pr(>F)
## HandWidth          1 343.86  343.86 13.3278    0.001023 **
## Gender             1 949.42  949.42 36.7985 0.000001329 ***
## HandWidth:Gender   1  77.23   77.23  2.9935    0.094229 .
## Residuals         29 748.21   25.80
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

This type of *sequential sum of squares* Analysis of Variance table should be getting fairly familiar to you now, but let's unpack what this means. There are four rows in the summary table - one for each of the terms in the model (`HandWidth`, `Gender` and `HandWidth:Gender`), and one for the `Residuals` (the unexplained variation that remains after fitting the model). The table includes degrees of freedom (`Df`), sum of squares (`Sum Sq`), mean sum of squares (`Mean Sq`) and the associated F and p-values (`F value` and `Pr(>F)`.

You can interpret the mean sum of squares column in terms of the amount of variation in the response variable (Height) that is explained by the term: The table first tells us the amount of variation (in terms of Mean Sum of Squares) in Height that is captured by a model that includes a common slope for both genders (881.6). Then it tells us that an *additional* bit of variation (471.13) is captured if we allow the intercepts to vary with gender. Then it tells us that a small additional amount of variation is explained by allowing the slope to vary between the genders (3.65). Finally, there is a bit of unexplained variation left over (Residuals) (26.29). So you can see that hand width explains most variation, followed by gender, followed by the interaction between them.

You would report from this table something like this:

Hand width and gender both explain a significant amount of the variation in height (ANCOVA - Handwidth: F =, d.f. = 1 and , p<0.001; Gender: , d.f. = 1 and , p<0.001). The interaction effect was not significant, which means that the slopes of the relationship between hand width and height are not significantly different (ANCOVA - , d.f. = 1 and , p = .

It is of course useful to take the interpretation a bit further. You could do this with reference to the plot - e.g. *Figure X shows the clear positive relationship between hand width and height and shows that the intercept for females is smaller than that for males. This which means that, for a given hand width, males tend to be taller.*

17.3 The summary of coefficients (`summary`)

To put some quantitative numbers on this description of the pattern we need to get the summary from R.

```
summary(mod_A)
```

```
##
```

```
## Call:
## lm(formula = Height ~ HandWidth + Gender + HandWidth:Gender,
##     data = classData)
##
## Residuals:
##     Min      1Q  Median      3Q     Max
## -9.0391 -4.0391 -0.6926  3.9288 11.4876
##
## Coefficients:
##                       Estimate Std. Error t value             Pr(>|t|)
## (Intercept)           160.7824     5.6546  28.434 < 0.0000000000000002 ***
## HandWidth               1.0321     0.6627   1.557              0.13024
## GenderMale             27.2210     8.9576   3.039              0.00499 **
## HandWidth:GenderMale  -1.6222     0.9376  -1.730              0.09423 .
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## Residual standard error: 5.079 on 29 degrees of freedom
## Multiple R-squared:  0.6469, Adjusted R-squared:  0.6103
## F-statistic: 17.71 on 3 and 29 DF,  p-value: 0.000001005
```

This summary table gives the coefficients of the statistical model, their standard errors, and the t-test results of whether the estimate is greater than 0. This is the same as the `summary` tables given for ANOVA and linear regression.

In the ANOVA `summary` tables, the estimates were given in relation to the *reference level* – the `(Intercept)` and these ANCOVA `summary` tables are no difference. Interpreting is best done with reference to the graph of the data and fitted model outputs (the graph above).

The reference level (the `(Intercept)`) is the intercept for the line for the first level of the categorical variable (Females, in this case). Here the model estimates that the intercept for Females is at 160.782 (i.e. if you extended the line out to the left it would eventually cross the y-axis at this point). The next coefficient `HandWidth` is the slope of this Female line (1.032). Then we have `GenderMale`: this coefficient is the difference in intercept between the Female and Male lines. This is followed by the intercept for the interaction term `HandWidth:GenderMale`: this is the difference between slopes for the two genders.

We can therefore do some simple arithmetic to get the equations (i.e. slopes and intercepts) of the lines for both genders. For females this is easy (they are reference level, so you can just read the values directly from the table) - the intercept is 160.782 and the slope is 1.032.

For males the intercept is 160.782 + 27.221 = 188.003. The slope is 1.032 + -1.622 = -0.590.

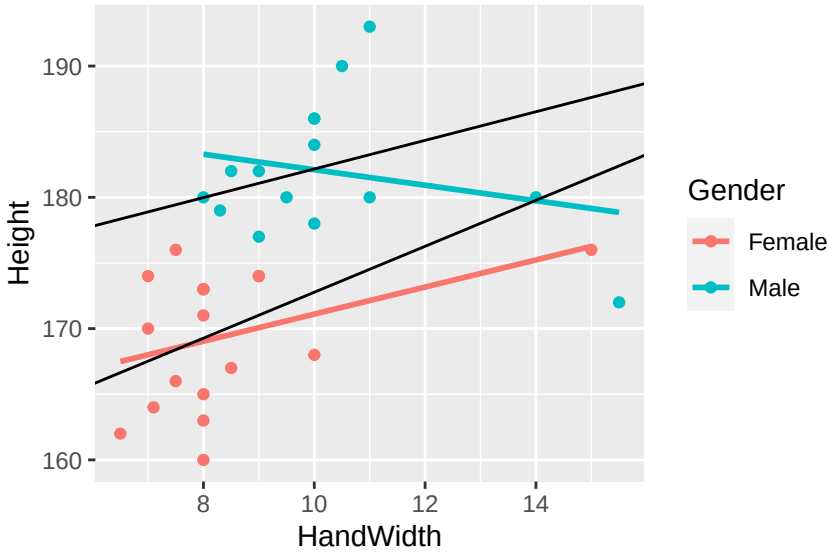
We could add these equations to our reporting of the results.

Figure X shows the clear positive relationship between hand width and height and shows that the intercept for females is smaller than that for males. This which means that, for a given hand width, males tend to be taller. The model

fit for males is Height = 1.75×HandWidth + 155.27 and the fit for females is Height = 1.09×HandWidth + 171.26

You could check these by using `geom_abline` to add lines with those equations to the plot (just as a "sanity check").

```
A +
  geom_abline(intercept = 155.27, slope = 1.75) +
  geom_abline(intercept = 171.26, slope = 1.09)
```



At the bottom of the `summary` output we are given the R^2 values. Because this model has several terms (i.e. variables) in it we should use the adjusted R^2 values. These have been corrected for the fact that the model has extra explanatory variables. So in this case, we could report that the model explains % of variation in Height (Adjusted R^2 = - not bad!

So, to describe this `summary` table more generally - the coefficients can be slopes, intercepts, differences between slopes, and differences between intercepts. They are slopes and intercepts for the first level of the categorical variable, and for the subsequent levels they are differences. Piecing these together can be hard to figure out without reference to the plot of the data and model fits - another good reason to plot your data!

17.4 Simplifying the model

Our results above showed that the interaction between the gender and hand width was not significant. Think about what that means? It means that the effect of hand width on height (the slope) does **not** depend on gender. Therefore, one can argue that we don't need to have a model that estimates both slopes - we could have a simpler model with one slope for both genders.

In fact, creating models that are as simple as possible to explain the observations is a useful goal that is captured by **the law of parsimony** or "*Occam's razor*", which essentially states that simple explanations for a phenomenon are favourable to complex explanations.

Let's refit the model without this non-significant interaction:

```
mod_B <- lm(Height ~ HandWidth + Gender, data = classData)
anova(mod_B)
```

```
## Analysis of Variance Table
##
## Response: Height
##           Df Sum Sq Mean Sq F value      Pr(>F)
## HandWidth  1 343.86  343.86  12.497    0.001345 **
## Gender     1 949.42  949.42  34.506 0.000001983 ***
## Residuals 30 825.45   27.51
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

Now all the terms in the model are significant.

```
summary(mod_B)
```

```
##
## Call:
## lm(formula = Height ~ HandWidth + Gender, data = classData)
##
## Residuals:
##      Min       1Q   Median       3Q      Max
## -11.0885  -3.2030   0.3518   4.1302  10.9086
##
## Coefficients:
##             Estimate Std. Error t value             Pr(>|t|)
## (Intercept) 167.5411     4.2221  39.682 < 0.0000000000000002 ***
## HandWidth     0.2216     0.4841   0.458                 0.65
## GenderMale   12.1128     2.0621   5.874           0.00000198 ***
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## Residual standard error: 5.245 on 30 degrees of freedom
## Multiple R-squared:  0.6104, Adjusted R-squared:  0.5844
## F-statistic:  23.5 on 2 and 30 DF,  p-value: 0.0000007231
```

The coefficient summary now gives us a two intercept estimates (152.0964 for females and 152.0964 + 2.1179 = 154.2143 for males) and single estimate for a slope that applies to both genders (2.1179).

Unfortunately, the handy `geom_smooth` function cannot handle this simpler model! We must take a slightly different, and sadly mode complicated approach:

What we need to do is **predict** using the model what the height will be under different conditions. Think of this as "plugging values into an equation".

We want to predict heights across the range of hand widths (from 6.5cm to 11cm), and we need to do this for males and females.

We do this by creating a "fake" dataset to predict from using the useful function `expand.grid`. This function takes inputs from columns of data and "expands" them to ensure that all possible combinations are included.

```
predictData <- expand.grid(
  HandWidth = c(6.5, 11),
  Gender = c("Male", "Female")
)
predictData
```

```
##   HandWidth Gender
## 1       6.5   Male
## 2      11.0   Male
## 3       6.5 Female
## 4      11.0 Female
```

Now we can use these values to `predict` what the heights will be for those particular combinations of values. The arguments for the `predict` function are the model name, then `newdata =` to give the function the data that you want to predict from. Here we can use the function to add the models predicted fitted value (`fit`) to the `predictData` object we just created.

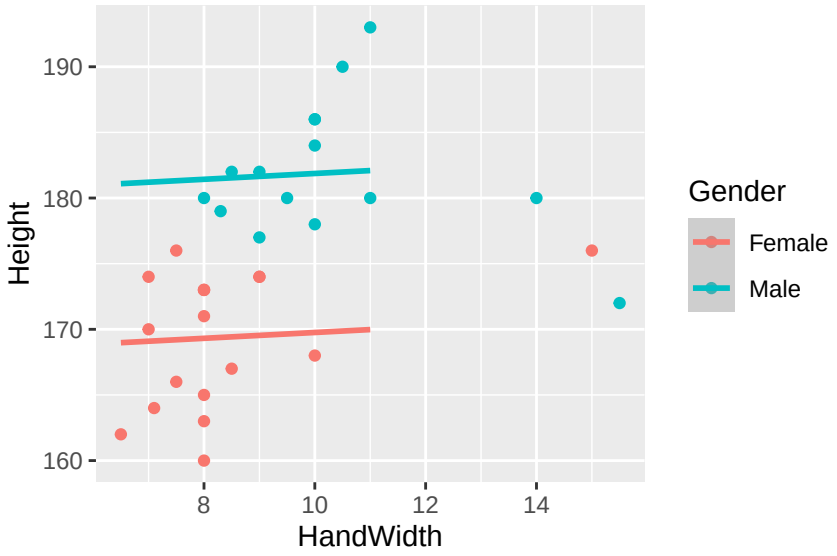
```
predictData$Height <- predict(mod_B, newdata = predictData)
predictData
```

```
##   HandWidth Gender   Height
## 1       6.5   Male 181.0943
## 2      11.0   Male 182.0914
## 3       6.5 Female 168.9814
## 4      11.0 Female 169.9786
```

Now we can add lines for these predicted values to our plot. We do this using the `geom_smooth` function as before, but this time we use the arguments `data = predictData` to tell R to use the new data, and `stat = "identity"` and to ensure that we plot the data rather than fitting any model.

You may wish to add an error ribbon to these lines. We will cover this in a later class (but see pages 159-164 in the GSWR textbook).

```
ggplot(classData, aes(
  x = HandWidth, y = Height,
  colour = Gender
)) +
  geom_point() +
  geom_smooth(data = predictData, stat = "identity")
```



We could report this in the usual way but first saying something like: *"The interaction term was not significant (F = xxxx, d.f. 1 and xx, p = XXX) and I therefore simplified the model to remove this term. The resulting model with just HandWidth and Gender …"*

Chapter 18

Linear models with >1 categorical explanatory variables

In the one-way ANOVA we covered in the previous chapter we were interested in understanding the effect of a single categorical explanatory variable with two or more levels on a continuous response variable. Although the explanatory variable must be categorical (i.e. with discrete levels), it could represent a continuous variable. For example, the explanatory variable could be a two-level soil nutrient level (high or low), even though nutrient level is a continuous variable and one could measure the actual quantitative value of nutrients in mg/g.

The two-way ANOVA is an extension of one-way ANOVA that allows you to investigate the effect of **two** categorical variables. This can be useful in an experimental context.

For example, one might have run an experiment investigating in the effect of two types of diet (*lowProtein* and *highProtein*), and genotype (*gt1* and *gt2*), on adult size of a pest species. It is worth thinking about what potential outcomes there are for this experiment. There may be no effect of diet, and no effect of genotype. There may be an effect of one of these variables but not the other. The effect of the diet might be the same for the different genotypes, or it might be different. Some of these different possible outcomes are illustrated in Figure 18.1. the titles indicate with Y (yes) or N (no) whether the figure shows a significant diet, genotype (gt) or interaction (int) effect. The dotted lines joining the estimates for the two genotypes are a kind of **interaction plot**: where they are parallel, there is no interaction.

In the model we aim to quantify these effects, and ask if they are statistically significant (i.e. if the effect sizes are >0). We divide the effects of the explanatory variables into two types: **main effects** and **interaction effects**. The main effects are the overall effect of the explanatory variables (genotype and diet in this case) while the interaction effect allows us to ask whether one main effect *depends on another*. In this case we are asking whether *the effect of diet depends*

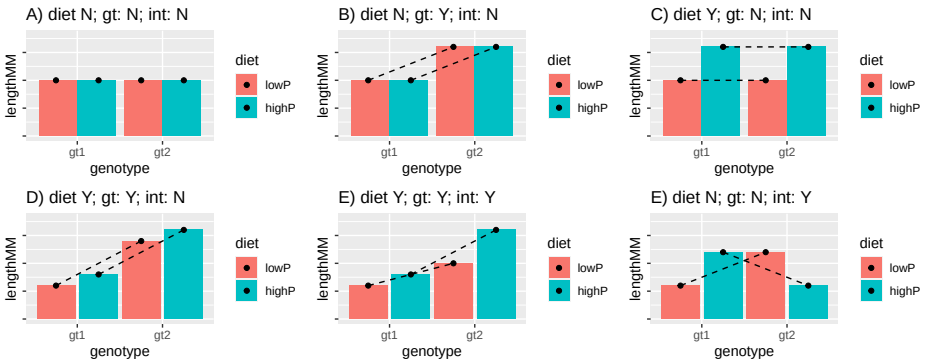


Figure 18.1: Some potential results of the experiment. There may be a significant effect (or not) of both of the main effects (diet and genotype) and there may be a significant interaction effect (or not).

on genotype (and vice versa). Make sure that you understand this important concept.

18.1 Fitting a two-way ANOVA model

Let's use R to fit a two-way ANOVA model using data from the example I just described. As with one-way ANOVA, you can fit a two-way ANOVA model in R using `lm`.

> Remember to load the `dplyr`, `magrittr` and `ggplot` packages, and to set your working directory correctly.

First, import the `insectDiet.csv` data and plot it, to produce a plot like in Figure 18.2. From looking at the graph in Figure 18.2 you can see (a) genotype 1 tends to be larger than genotype 2; (b) insects raised on a high protein diet tend to be larger than those on a low protein diet; and (c) the effect of the diet (i.e. the *difference* in size between the insects raised on the different diets) is larger for genotype 1 than it is for genotype 2. But are these differences statistically meaningful?

```
insectDiet <- read.csv("CourseData/insectDiet.csv")

ggplot(insectDiet, aes(x = genotype, y = lengthMM, fill = diet)) +
  geom_boxplot() +
  xlab("Genotype") +
  ylab("Length (mm)")
```

To address this question, we will fit a linear model (the two-way ANOVA) to estimate the effects of diet and genotype.

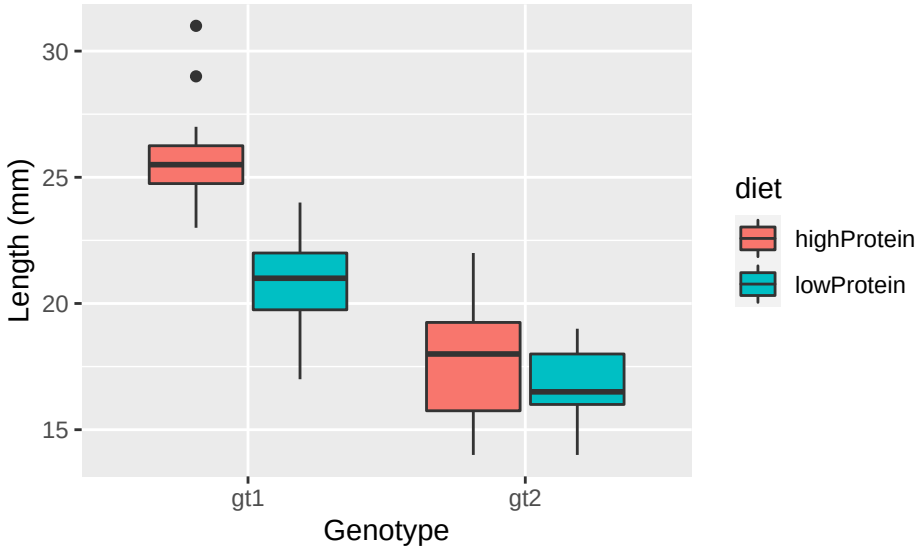


Figure 18.2: The effect of diet protein content and genotype on adult size of an insect species

The model formula is `lengthMM ~ genotype + diet + genotype:diet`.

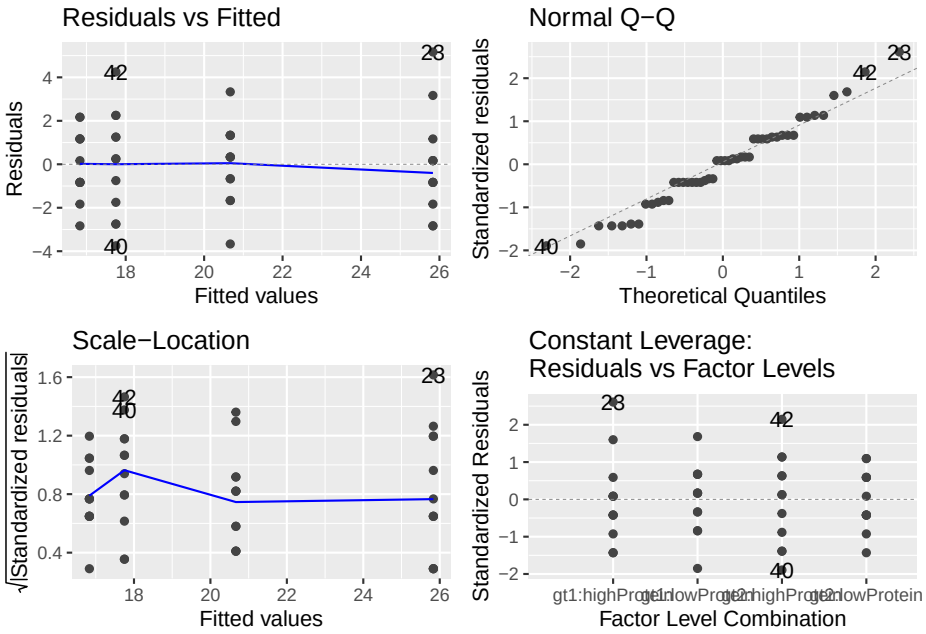
Let's try to understand this. The `genotype + diet` part represents the **main effects** of these two variables, and the `genotype:diet` part represents the **interaction effect** between them. This formula*can* be shortened to `lengthMM ~ genotype * diet` (i.e. this is exactly equivalent to the more complicated-looking formula), but I recommend to use the longer version because it is clearer.

So we fit the model like this - putting the formula first, then telling R which data to use:

```
mod_A <- lm(lengthMM ~ genotype + diet + genotype:diet,
  data = insectDiet
)
```

Then we can look at diagnostic plots, as with ANOVA etc.:

```
library(ggfortify)
autoplot(mod_A)
```



These all look OK. The slightly odd structure in the QQ-plot is caused by the fact that the length data are rounded to the nearest millimeter. There is no evidence of heteroscedasticity (left hand plots) now any major outliers.

18.2 Summarising the model (`anova`)

Since we are satisfied with the diagnostic plots we can proceed by summarising the model using first `anova` and then `summary`.

```
anova(mod_A)
```

```
## Analysis of Variance Table
##
## Response: lengthMM
##               Df Sum Sq Mean Sq F value             Pr(>F)
## genotype       1 426.02  426.02  99.575 0.0000000000007135 ***
## diet           1 111.02  111.02  25.949 0.0000070636935073 ***
## genotype:diet  1  54.19   54.19  12.665          0.0009073 ***
## Residuals     44 188.25    4.28
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

This summary `Analysis of Variance Table` is similar to the ones you have already seen for one-way ANOVA and linear regression. It just has some extra rows because you have extra explanatory variables. It shows you the degrees of freedom for the different terms in the model (all 1, because they have two

levels), the sum of squares (`Sum Sq`) and mean sum of squares (`Mean Sq`) and the associated `F value` and p-value (`Pr(>F)`). Those F values are all large, leading to highly-significant p-values.

This means that all of those terms in the model explain a significant proportion of the variation in insect length.

But as you know, this summary table doesn't tell you the direction of the effects. The obvious way to understand your data is to simply look at the plot you have already produced. You could also make an **interaction plot** which is a simplified version of the plot of the raw data.

To do this you first need to create a summary table using `dplyr` tools `summarise` and `group_by` to get the mean and standard errors of the mean:

```
insectDiet_means <-
  insectDiet %>%
  group_by(genotype, diet) %>% # <- remember to group by *both* factors
  summarise(MeanLength = mean(lengthMM), SELength = sd(lengthMM) / sqrt(n()))
insectDiet_means
```

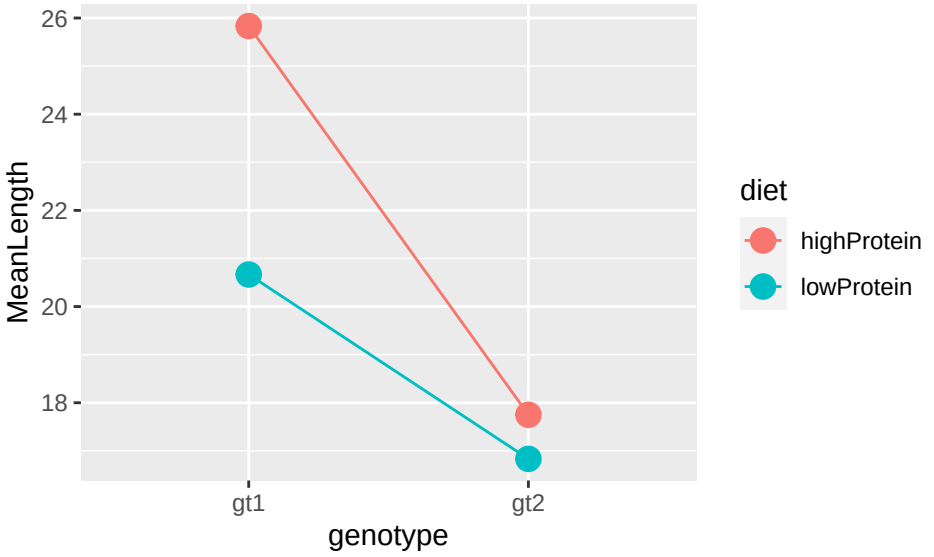
```
## # A tibble: 4 x 4
## # Groups:   genotype [2]
##   genotype diet        MeanLength SELength
##   <chr>    <chr>            <dbl>    <dbl>
## 1 gt1      highProtein       25.8    0.672
## 2 gt1      lowProtein        20.7    0.527
## 3 gt2      highProtein       17.8    0.698
## 4 gt2      lowProtein        16.8    0.458
```

Then you can make a simple plot of this information by plotting points, and lines joining them:

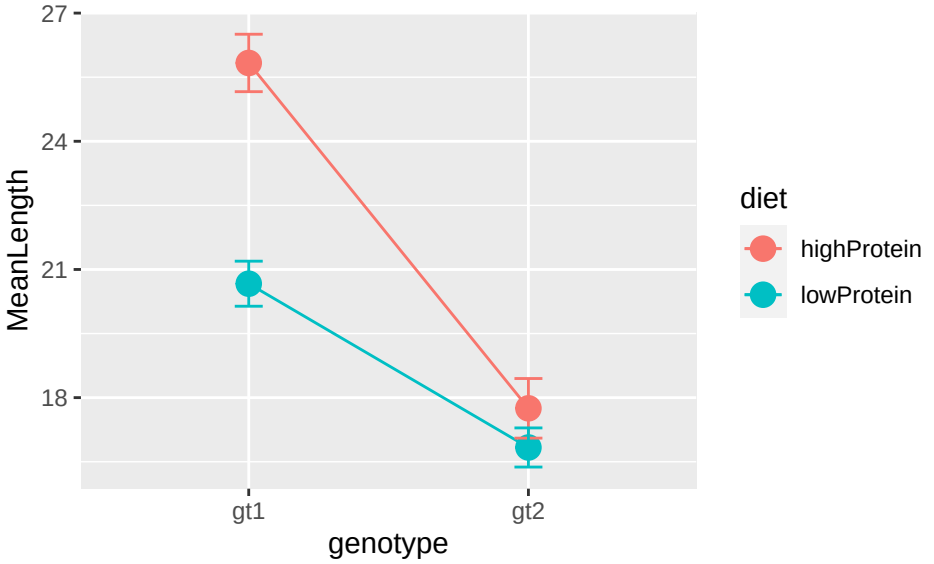
```
(A <- ggplot(
  insectDiet_means,
  aes(x = genotype, y = MeanLength, colour = diet, group = diet)
) +
  geom_point(size = 4) +
  geom_line())
```

You could add error bars to the points by adding a line defining the `ymin` and `ymax` values from the data summary like this:

```
ggplot(
  insectDiet_means,
  aes(
    x = genotype, y = MeanLength, colour = diet, group = diet,
```




```
    ymin = MeanLength - SELength, ymax = MeanLength + SELength
  )
) +
  geom_point(size = 4) +
  geom_line() +
  geom_errorbar(width = 0.1)
```



But are these points statistically significantly different from each other? To answer that question we need to use a post-hoc test

```
library(agricolae)
HSD.test(mod_A, trt = c("diet", "genotype"), console = TRUE)
```

```
##
## Study: mod_A ~ c("diet", "genotype")
##
## HSD Test for lengthMM
##
## Mean Square Error:  4.278409
##
## diet:genotype,  means
##
##                 lengthMM      std  r Min Max
## highProtein:gt1 25.83333 2.329000 12  23  31
## highProtein:gt2 17.75000 2.416797 12  14  22
## lowProtein:gt1  20.66667 1.825742 12  17  24
## lowProtein:gt2  16.83333 1.585923 12  14  19
##
## Alpha: 0.05 ; DF Error: 44
## Critical Value of Studentized Range: 3.775958
##
## Minimun Significant Difference: 2.254643
##
## Treatments with the same letter are not significantly different.
##
##                 lengthMM groups
## highProtein:gt1 25.83333      a
## lowProtein:gt1  20.66667      b
## highProtein:gt2 17.75000      c
## lowProtein:gt2  16.83333      c
```

The important part of this output is at the bottom where it tells us `Treatments with the same letter are not significantly different.`. You can see that the mean lengths between diets for genotype 1 are significantly different (they do not share a letter). However, there is no significant difference between diets for genotype 2 (they share the same letter, `c`). The two genotypes are also significantly different from each other.

18.3 Summarising the model (`summary`)

This (above) is generally enough information for a complete write up of results. However, you can ask R to provide the model summary that includes the R^2 values, coefficient estimates and standard errors using `summary`.

```
summary(mod_A)
```

```
##
## Call:
## lm(formula = lengthMM ~ genotype + diet + genotype:diet, data = insectDiet)
##
## Residuals:
##     Min      1Q  Median      3Q     Max
## -3.7500 -1.0417  0.1667  1.2500  5.1667
##
## Coefficients:
##                            Estimate Std. Error t value             Pr(>|t|)
## (Intercept)                 25.8333     0.5971  43.264 < 0.0000000000000002 ***
## genotypegt2                 -8.0833     0.8444  -9.572     0.00000000000253 ***
## dietlowProtein              -5.1667     0.8444  -6.118     0.00000022595031 ***
## genotypegt2:dietlowProtein   4.2500     1.1942   3.559             0.000907 ***
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## Residual standard error: 2.068 on 44 degrees of freedom
## Multiple R-squared:  0.7585, Adjusted R-squared:  0.742
## F-statistic: 46.06 on 3 and 44 DF,  p-value: 0.0000000000001254
```

The most useful thing shown here is the R^2 value. Because we have several terms in the model we should use the `Adjusted R-squared` value of 0.742. This indicates that our model explains 74.2% of variation in insect length.

The next bit is not 100% necessary most of the time…

We already have a good idea of the mean values and standard errors for these data look because we calculated them above directly from the data. For completeness though I will now run through the coefficient estimates part of the summary table.

The coefficient `Estimates` here are interpreted in a similar way to a one-way ANOVA. Again, it is important to know what the reference point is. When you understand this you can reconstruct the mean values for the various levels of the variables that are estimated by the model. You will see that the model estimates lead to precisely the same estimates as obtained from summarising the data.

Here you can see that:

- The `(Intercept)` is 25.8333 and must refer to the point for *genotype 1* on a *high protein diet* (look at the value of the intercept and compare to the graph/summary table, and/or the output from the Tukey test).
- The second coefficient (`genotypegt2`) is -8.0833 which is the **difference** between the reference (intercept) and the value for *genotype 2* on a *high protein diet*: (25.8333 + (-8.0833) = 17.75).

- The third coefficient (`dietlowProtein`) is -5.1667 which is the difference between the reference point and for *genotype 1* on a *low protein diet*: (25.8333 + (-5.1667) = 20.6666).
- The final coefficient `dietlowProtein:genotypegt2` is 4.25 and is "interaction effect" of diet and genotype and represents the *additional* effect of genotype when it is on diet. In other words, in comparison to the reference point (genotype 1 & high protein diet), the effect of a low protein diet is negative (-8.0833), as is the effect of being genotype 2 (-5.1667). However, having both a low protein diet **and** being genotype 2 leads to an additional positive effect (4.25) on length. The resulting estimate of mean length for *genotype 2* on a *low protein diet* is 25.8333 + (-8.0833) + (-5.1667) + 4.25 = 16.833.

This is a bit complicated so my advice is generally to refer to the figures and the outputs of the `Tukey.HSD` function to obtain the estimate in the different groups.

The logic and methods of the two-way ANOVA can be extended to produce n-way ANOVA with n categorical variables.

18.4 Exercise: Fish behaviour

Individual differences in animal personality and external appearance such as colouration patterns have both been extensively studied separately. A significant body of research has explored many of pertinent ecological and biological aspects that can be affected by them and their impact upon fitness. Currently little is known about how both factors interact and their effect on reproductive success.

Researchers carried out a study looking at differences in personality and its interaction with colour phenotype in zebra fish (*Danio rerio*). They used two colour morphs, "homogeneous" which has clearly defined lateral stripes, and "heterogeneous" which has more variable and less clear patterns.

They also assigned individuals to two personality types which they called "Proactive" (adventurous, risk taking) and "Reactive" (timid, less risk taking). They did this by recording how they explore a new environment

The two variables of interest are:

- Colour pattern (homogeneous and heterogeneous)
- Personality (proactive and reactive)

The research questions are: *What is the relative influence of colour pattern and personality? Which is more important? How do the variables interact to determine fitness? e.g. do proactive individuals do better than reactive ones, and does this depend on colour pattern? Or some other pattern?*

1) Import the data set, `fishPersonality.csv`

2) Plot the data (e.g. as a box plot)

3) Fit an ANOVA model using `lm`.

4) Look at diagnostic plots from this model (`autoplot`)

5) Use `anova` to get an Analysis of Variance summary table, and interpret the results.

6) Get the coefficient summary (`summary`) and interpret the output.

7) Do post-hoc Tukey tests (e.g. using `HSD.test` from the `agricolae` package). Interpret the results.

8) Sum up your findings with reference to the initial research questions.

Chapter 19

Generalised linear models

The models we have covered so far are ordinary linear models (including ANOVA, ANCOVA, ordinary linear regression etc.) that assume that the relationship between the explanatory variables and the response variable is linear, and that the systematic error in the model is constant (homoscedastic, i.e. the standard deviation of the data does not depend on the magnitude of the explanatory variable).

In many cases this will not be the case. Non-linearity and heteroscedasticity tend to go hand-in-hand. Sometimes, for example, the data show an exponential growth type of pattern, and/or may be bounded in such a way that the errors cannot be homoscedastic. For example, counts of number of species on islands of different sizes have a lower bound at zero (you can't have negative numbers of species!) and increase exponentially while the standard deviations are are small for small islands and large for large islands.; ratio data or percentage data such as proportion of individuals dying/surviving is bounded between 0 and 1 (0 - 100%).

Transformation of the response variable could an option to linearise these data (although there would be problems with 0 values (because log(0) = -Infinity)), but a second problem is that the ordinary linear model assumes "homoscedasticity" - that the errors in the model are evenly distributed along the explanatory variables. This assumption will be violated in most cases. For example, with count data (e.g. number of species in relation to island size), the errors for very small islands will be smaller than those for large islands. In fact, even if we transform the response variable, for example by log transformation, the predictions of the model will allow errors that include negative counts. This is clearly a problem!

Generalised linear models (GLMs) solve these problems by not only applying a transformation but also explicitly altering the error assumption. They do this using a **link function** to carry out the transformation and by choosing an **error structure** (sometimes referred to as **family**, or **variance function**). The choice of link and error structure can be a bit confusing, but there are so-called "canonical links" that are commonly associated with particular error structures. For example, a model for count data would usually have a log link

and a Poisson error structure.

The flexibility of the GLM approach means that one can fit GLM versions of all of the models you have already learned about until this point: ANOVA-like GLMs, ANCOVA-like GLMs, ordinary regression-like GLMs and so on.

In this chapter I will focus on this count data and in the next chapter I will broaden the focus to illustrate uses of other data types.

19.1 Count data with Poisson errors.

The most common kind of count data where Poisson errors would be expected are frequencies of an event: we know how many times an event happened, but not how many times it did not happen (e.g. births, deaths, lightning strikes).

In these cases:

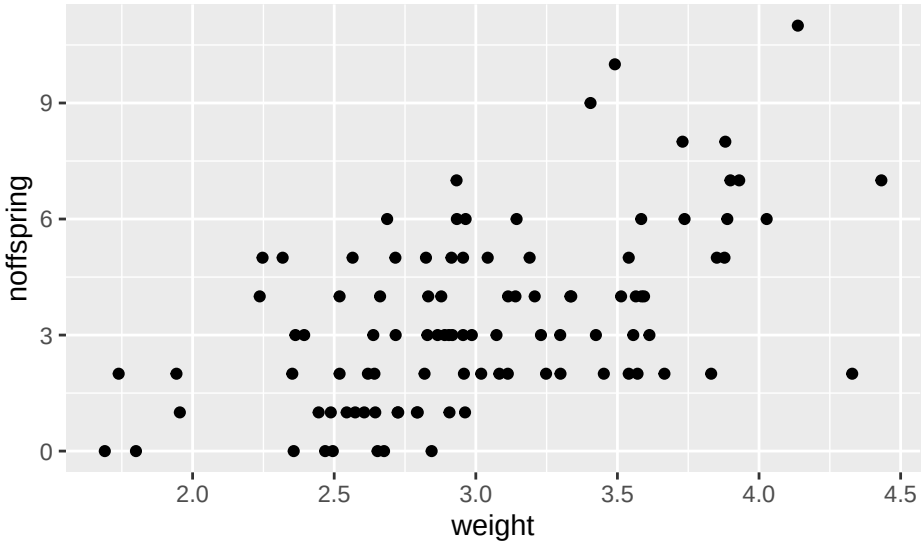
- Linear model could lead to negative counts.
- Variance of response likely to increase with mean (it usually does with count data).
- Errors are non-normal.
- Zeros difficult to deal with by transformation (e.g. log(0) = -Inf).
- Other error families do not allow zero values.

The standard ("canonical") link used with the Poisson error family is the log link. The log link ensures that all fitted (i.e. predicted) values are positive, while the Poisson errors take account of the fact that the data are integer and the variance scales 1:1 with the mean (i.e. variance increases linearly and is equal to the mean). There are other potential link and error families that *could* be used with this kind of data, but we'll stick with the standard ones here. Lets look at a couple of examples…

19.1.1 Example: Number of offspring in foxes.

This example uses the `fox.csv` data set. This data set gives the number of offspring produced by a group of foxes, alongside the weight (in kg) of the mothers. Let's import and plot the data.

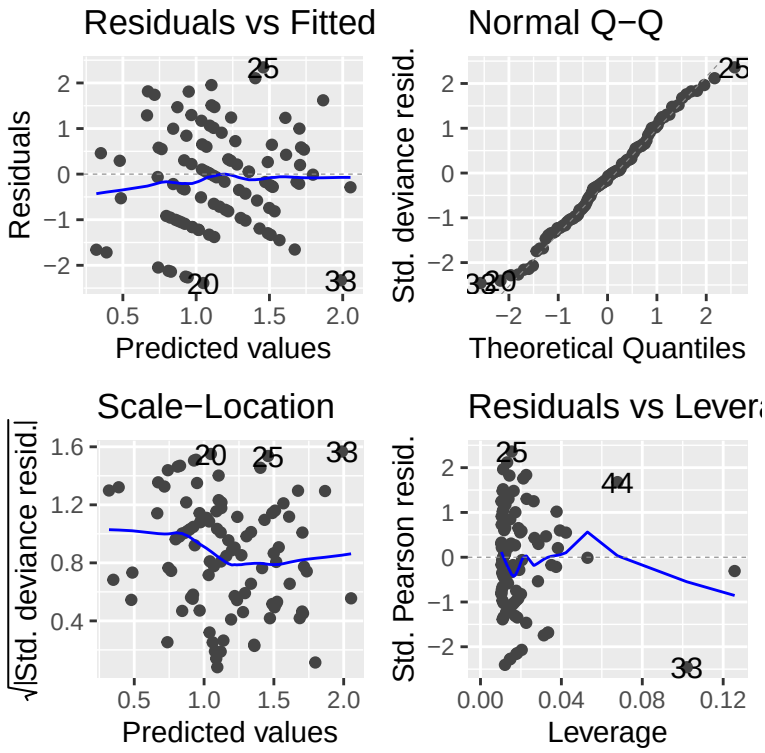
> Remember to load the `dplyr`, `magrittr` and `ggplot` packages, and to set your working directory correctly.



The first thing to notice is that, like all count data, the data are formed into horizontal rows of data reflecting the fact that the response data are integer values. There is clearly an increasing pattern, but how can we formally test for a statistical relationship. It is obvious that fitting an ordinary linear model though this data would not be the right approach: this would lead to the prediction of negative number of offspring for small foxes, and also, the variance appears to increase with weight/number of offspring. Therefore this is a good candidate for a GLM. The data are bounded at 0, and are integer values, and for this reason the usual approach would be to fit a GLM with Poisson errors (and the standard log link).

```
mod1 <- glm(noffspring ~ weight, data = fox, family = poisson)
```

After fitting the model it is a good idea to look at the model diagnostics, using `autoplot` from the `ggfortify` package.



These diagnostic plots are, basically, the same as the ones you saw for `lm` models. There is one difference and that is that the plots use something called standardised *deviance* residuals instead of just standardised residuals. These are transformed versions of the residuals that will look *normal* (in the statistical sense) if the family of hte GLM is appropriate. So, if "poisson" is the right family, the QQ-plot should show points on the diagonal dashed line, and the Scale-location plot should have no strong patterns.

Now we can ask for the Analysis of Variance table for this model. This is exactly the same procedure as for the previous linear models (ANOVA, ANCOVA etc.) except for GLMs one must also specify that you would like to see the results of significance tests using the `test = "F"` or `test = "Chi"`. For Poisson and binomial GLMs the chi-squared test is most appropriate while for Gaussian (normal), quasibinomial and quasipoisson models the F test is most appropriate.

```
anova(mod1, test = "Chi")
```

```
## Analysis of Deviance Table
##
## Model: poisson, link: log
##
## Response: noffspring
##
## Terms added sequentially (first to last)
```

```
##
##
##        Df Deviance Resid. Df Resid. Dev         Pr(>Chi)
## NULL                      99     166.84
## weight  1   44.124        98     122.72 0.00000000003082 ***
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

This summary table tells us that the single explanatory variable (`weight`) is fantastically important (p-value is very small indeed).

We can then ask for the coefficient summary using `summary`.

```
summary(mod1)
```

```
##
## Call:
## glm(formula = noffspring ~ weight, family = poisson, data = fox)
##
## Deviance Residuals:
##     Min       1Q   Median       3Q      Max
## -2.3891  -0.9719  -0.1183   0.5897   2.3426
##
## Coefficients:
##             Estimate Std. Error z value        Pr(>|z|)
## (Intercept) -0.74981    0.31107  -2.410          0.0159 *
## weight       0.63239    0.09502   6.655 0.0000000000283 ***
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## (Dispersion parameter for poisson family taken to be 1)
##
##     Null deviance: 166.85  on 99  degrees of freedom
## Residual deviance: 122.72  on 98  degrees of freedom
## AIC: 405.56
##
## Number of Fisher Scoring iterations: 5
```

> GLM model coefficients and predicted values, are expressed on the scale of the *linear predictor* (i.e. the transformed data scale). It is usually desirable to "backtransform" to the natural scale before plotting. See below.

The model coefficients and their standard errors are given on the scale of the linear predictor. They tell us that there is a significant association between the weight of the fox mother and the number of offspring she will produce: larger foxes produce more offspring. Because the coefficients are given on the scale of

the linear predictor rather than on the real scale it is useful to plot predictions of the model to visualise the relationship.

To do that we must (1) tell the model what to predict `from` i.e. we must provide a suitable sequence of numbers to predict from using `seq`, (2) use the `predict` function to predict values (`fit`) from the model. We use the argument `type = "response"` to tell the function that we want the predictions on the back-transformed (real) scale rather than on the scale of the linear predictor. We add the argument `se.fit = TRUE` to tell the function to give us the standard error estimates of the fit. The `se.fit` values are added or subtracted from the fit to obtain the plus/minus standard errors. We can multiply these by 1.96 to get the 95% confidence intervals of the fitted values.

```
# Vector to predict from
newData <- data.frame(weight = seq(1.7, 4.4, 0.01))

# Predicted values (and SE)
predVals <- predict(mod1, newData,
  type = "response",
  se.fit = TRUE
)

# Create new data for the predicted fit line
newData <- newData %>%
  mutate(noffspring = predVals$fit) %>%
  mutate(ymin = predVals$fit - 1.96 * predVals$se.fit) %>%
  mutate(ymax = predVals$fit + 1.96 * predVals$se.fit)
```

Take a look at this data to make sure it looks OK.

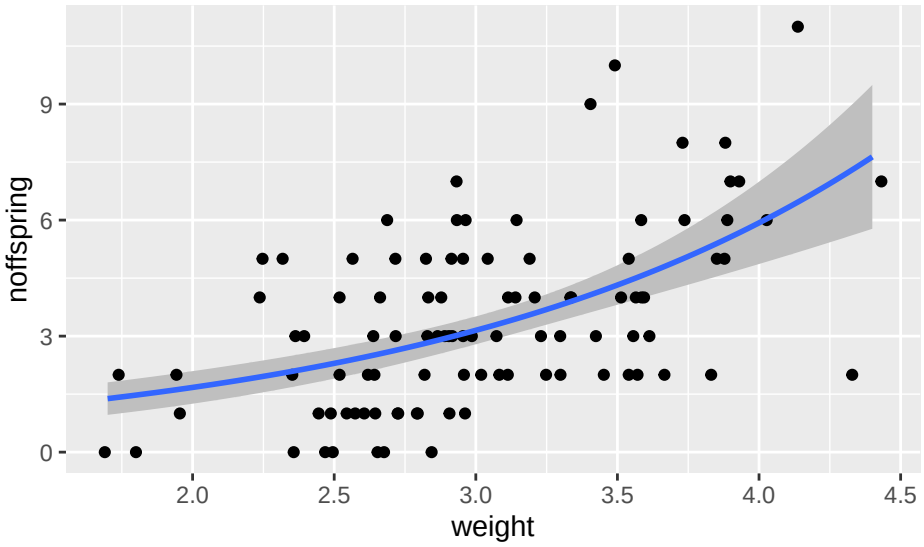
```
head(newData)
```

```
##   weight noffspring      ymin     ymax
## 1   1.70   1.384392 0.9649333 1.803850
## 2   1.71   1.393174 0.9734837 1.812865
## 3   1.72   1.402013 0.9821017 1.821924
## 4   1.73   1.410907 0.9907879 1.831026
## 5   1.74   1.419858 0.9995426 1.840173
## 6   1.75   1.428865 1.0083663 1.849364
```

This looks OK. Now we can plot the data and add a the model fit line, and a "ribbon" representing the errors (the 95% confidence interval for the line).

```
(A <- ggplot(fox, aes(x = weight, y = noffspring)) +
  geom_ribbon(data = newData, aes(
    x = weight, ymin = ymin,
```

```
    ymax = ymax
  ), fill = "grey75") +
  geom_point() +
  geom_smooth(data = newData, stat = "identity"))
```



So we could summarise this something like this:

Methods: *I modelled the association between mother's weight and number of pups produced using a generalised linear model with a log link and Poisson error structure. This is appropriate because the data are count data (number of pups) that are bounded at 0 with increasing variance with increased maternal weight.*

Results: *The GLM showed that maternal weight was significantly associated with the number of pups produced (GLM: Null Deviance = 166.8, Residual Deviance = 122.7, d.f. = 1 and 98, p <0.001). The slope of the relationship was 0.63 (on the log scale). The equation of the best fit line was log(nOffspring) = -0.75 + 0.63 × MotherWeight (see Figure XXX)*

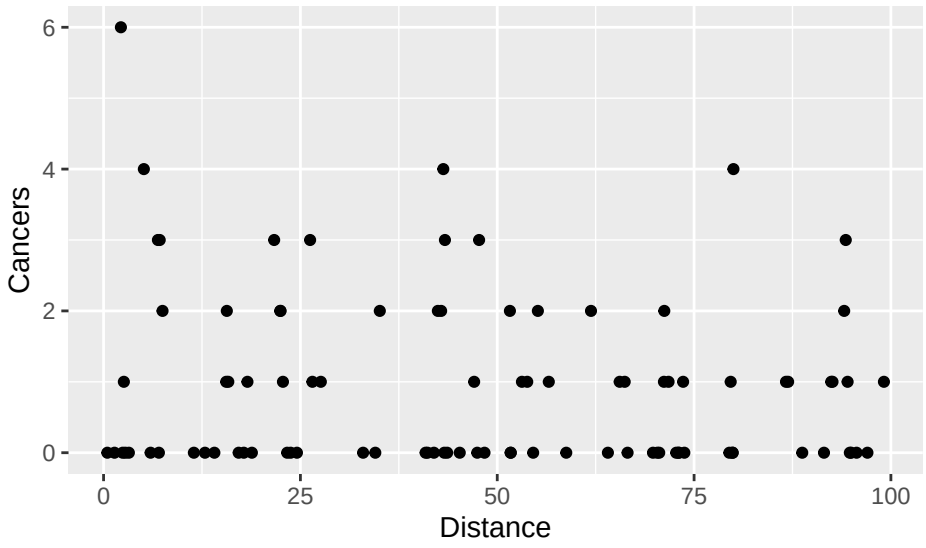
19.1.2 Example: Cancer clusters

This data show counts of prostate cancer and distance from a nuclear processing plant. Lets take a look at the data.

Let's first import the data (`cancer.csv`) and use summary to examine it by plotting it:

First we can see that there are no negative count values.

```
cancer <- read.csv("CourseData/cancer.csv")

ggplot(cancer, aes(x = Distance, y = Cancers)) +
  geom_point()
```

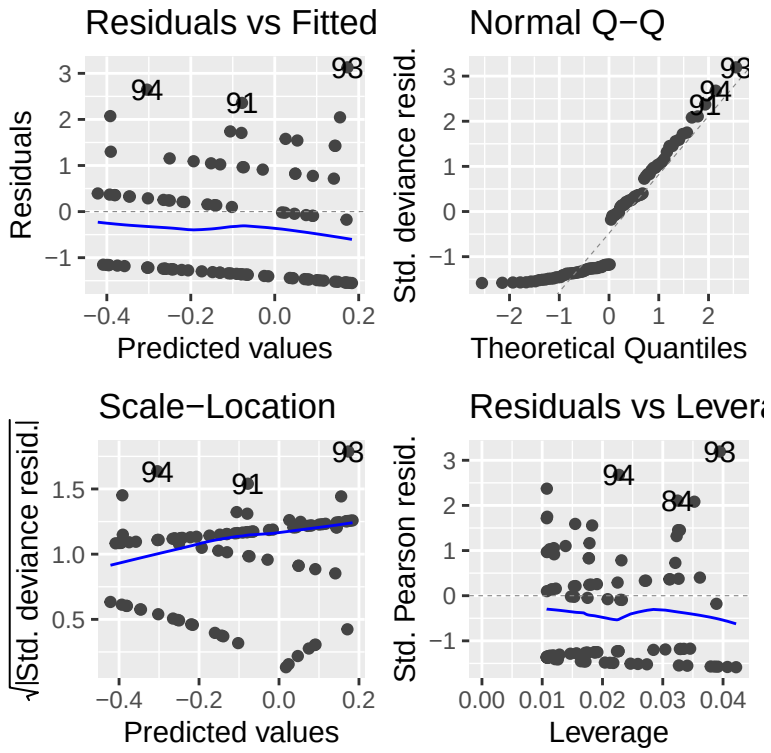


Again, you will notice that the data are formed into horizontal rows of integer response values. There are lots of zero values at all distances, but the biggest cluster (6 cases), is very close to the plant. But is there a relationship between the distance from the nuclear plant and the number of cancers?

Let's fit a Generalised Linear Model to find out. As before will assume that the error is Poisson (that they variance increases directly in proportion to the mean), and we will use the standard log link to ensure that we don't predict negative values:

```
mod1 <- glm(Cancers ~ Distance, data = cancer, family = poisson)
```

Next, plot the diagnostic plots.



These look a bit dodgy, but we'll stick with it for the moment.

Next ask for the Analysis of Variance table.

```
anova(mod1, test = "Chi")
```

```
## Analysis of Deviance Table
##
## Model: poisson, link: log
##
## Response: Cancers
##
## Terms added sequentially (first to last)
##
##
##          Df Deviance Resid. Df Resid. Dev Pr(>Chi)
## NULL                        93     149.48
## Distance  1   2.8408        92     146.64   0.0919 .
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

The ANOVA table tells us that there is no significant effect of the Distance variable. In other words a model that includes the `Distance` term does not explain significantly more variation than the `NULL` model that includes no terms

and instead assumes that variation in cancer incidence is simply caused by random variation.

We needn't go further with this model, but go ahead and plot the model in any case (just for practice).

```
summary(mod1)
```

```
##
## Call:
## glm(formula = Cancers ~ Distance, family = poisson, data = cancer)
##
## Deviance Residuals:
##     Min       1Q   Median       3Q      Max
## -1.5504  -1.3491  -1.1553   0.3877   3.1304
##
## Coefficients:
##              Estimate Std. Error z value Pr(>|z|)
## (Intercept)  0.186865   0.188728   0.990   0.3221
## Distance    -0.006138   0.003667  -1.674   0.0941 .
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## (Dispersion parameter for poisson family taken to be 1)
##
##     Null deviance: 149.48  on 93  degrees of freedom
## Residual deviance: 146.64  on 92  degrees of freedom
## AIC: 262.41
##
## Number of Fisher Scoring iterations: 5
```

Use the approach from the fox example as guidance to make a plot with a fit line.

19.2 Exercise: Maze runner

In an experiment, researchers studied the ability of children and adults to navigate through a maze. They recorded the number of mistakes each person made before successfully completing the maze. The data (`maze.csv`) has two columns: `Age` (a categorical variable with two levels - Adult and Child) and `nErrors` a count of the number of errors that each subject makes.

In this example, you will be fitting a GLM equivalent of a t-test that is appropriate for count data.

1) Import the data and graph it (`geom_boxplot`). Try adding the points to the `ggplot` using the new (to you) function `geom_dotplot(binaxis = "y", stackdir = "center")`.

2) Fit an appropriate GLM.
3) Examine the diagnostic plots of the model (`autoplot`).
4) Get the analysis of variance (deviance) table (`anova`). What does this tell you?
5) Obtain the `summary` table. What does this tell you?
6) Use the coefficient information in the `summary` table to get the model predictions for average number of mistakes (plus/minus 95% Confidence interval). Remember that (i) the model summary is on the scale of the linear predictor, and (ii) the 95% CI can be calculated as 1.96 times the standard error. You can do these calculations "by hand", or using the `predict` function. Ask for help if you get stuck.

Chapter 20

Extending use cases of GLM

In the previous chapter we used the case of modelling count data, which is bounded at 0 and takes integer values, to understand how Generalised Linear Models work. In this chapter we extend our understanding by looking at another type of data, namely *binomial* data.

20.1 Binomial response data

There are three types of Binomial data, but all three types have the idea of "success" and "failure" at their heart. It is up to you, the modeller, to decide what these successes and failures are, but they can be any kind of data that can be coerced into two discrete categories. Common examples include pass/fail, survived/died, presence/absence and yes/no.

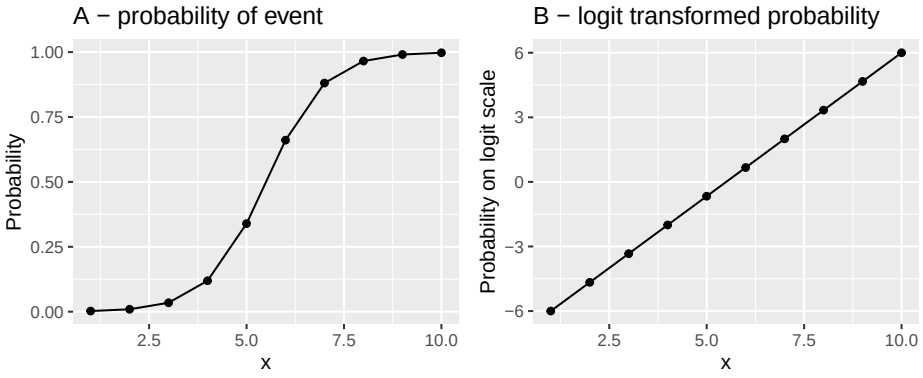
The three data types are as follows:

1) The data can be in the form of a two-level factor - often coded as zeros and ones (0/1 data). For example, this could represent whether an died (0) or survived (1) in a study; or it could represent male (`M`) or female (`F`) in a study trying to predict sex from other variables (e.g. size or morphology).
2) The data could be expressed as a numeric vector of proportions, bounded between 0 and 1 (i.e. can take any value between 0 and 1). For example these data could be percentages expressed as proportion, such as proportion (or percent) of individuals in a group surviving. In this case, the total number of cases that contributed to the proportion can be optionally included as `weights` in the regression model [1].
3) Finally, the data could be expressed as numbers in a two column matrix where the first column gives the number of "successes" and the second column gives the number of "failures".

[1] weights allow some data points to be more influential than others - for example you would want to give more importance to points that represent large sample sizes

The aim of the GLM is usually to estimate probability of "success" (e.g. survival, passing, scoring a goal…). Thus, the outcome (the predicted response, or fit, of the model) is a value between 0 and 1 on the natural scale, that can be interpreted as a probability (e.g. of survival, of success, of presence etc.)

The link function used for a binomial GLM is usually the *logit* transformation, and therefore another name for this type of regression is **logistic regression**. The logit transformation linearises an S-shaped curve and therefore allows a linear regression line to be fitted from data that follows this pattern. The function is $y = log(\frac{p}{1-p})$, where p is the probability or proportion. The inverse logit (the anti-logit) is $p = \frac{exp(y)}{exp(y)+1}$.



We **could** linearise the data and then fit an ordinary linear model using `lm` but (like with the Poisson regression) the other assumption of the ordinary linear model, homoscedasticity, would cause problems. With S-shaped binomial relationships the expected variance is small at the two ends of the range and large in the middle of the range. This contrasts strongly with the constant variance assumption of ordinary linear models. Therefore it is wise to account for this using a Generalised Linear Model that explicitly accounts for this variance structure.

Let's try a couple of examples.

> Remember to load the `dplyr`, `magrittr` and `ggplot` packages, and to set your working directory correctly.

20.2 Example: NFL field goals

In this example you will be dealing with binary data (0/1, failure/success) from the American National Football League (NFL). The data are a record of field goals, which are a relatively rare method of scoring where someone kicks the ball through over the crossbar and through the uprights of the goal during play.

Our aim is to estimate how the probability of success changes with distance from the goal. We already have a good idea that success probability will decline

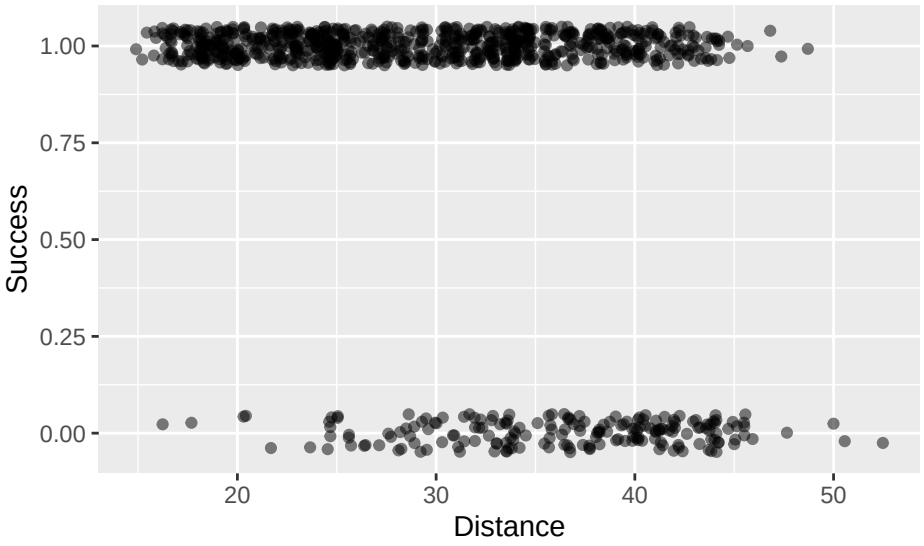
with increasing distance! But at what rate does the probability decline? And at what distance is there a probability of 0.5 (i.e. 50% chance of success)?

First, import the data and convert the distance from yards to meters.

```
NFL <- read.csv("CourseData/NFLfieldgoal.csv") %>%
  mutate(Distance = Distance * 0.9144)
```

Next, plot the data with `ggplot` (`geom_jitter` would be a good option, but you might like to adjust the `height` and `alpha` arguments. e.g. `geom_jitter(height = 0.05,alpha = 0.5)`). You can see that the data is distributed in two bands with 1 representing success and 0 representing failure.

```
(A <- ggplot(NFL, aes(x = Distance, y = Success)) +
  geom_jitter(height = 0.05, alpha = 0.5))
```



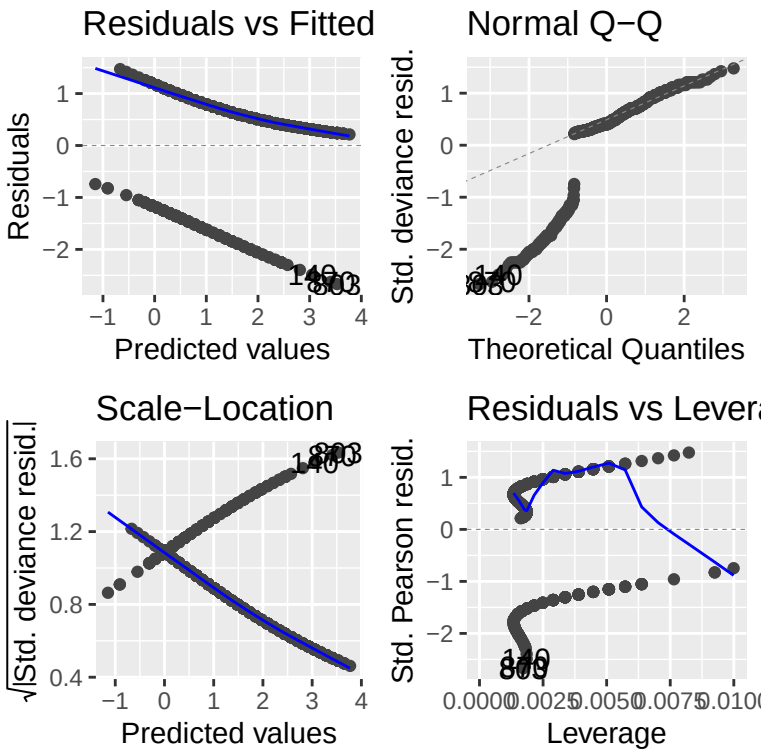
Now we can fit a GLM using an appropriate binomial error structure.

```
nflMod <- glm(Success ~ Distance, data = NFL, family = binomial)
```

As usual, we should look at the model diagnostics. These look pretty bad. One reason that these diagnostics (e.g. the QQ plot) can look bad is when we are missing important variables in the model. In this case, for example, the distribution could look "off" because we don't include important information on other aspects of the game (e.g. was the team winning or losing at the time? how many minutes until the end of the game? how experienced is the player?).

We can be fairly sure that `binomial` is the most appropriate variance structure because of the nature of the 0/1 data so let's stick with it for now!

```
library(ggfortify)
autoplot(nflMod)
```



We proceed in the normal way by obtaining the ANOVA table for the model. We need to specify that we want to calculated p-values using a "Chi" squared test. This shows us that indeed distance is an important variable in determining probability of success (not so surprising!)

```
anova(nflMod, test = "Chi")
```

```
## Analysis of Deviance Table
##
## Model: binomial, link: logit
##
## Response: Success
##
## Terms added sequentially (first to last)
##
##
```

```
##          Df Deviance Resid. Df Resid. Dev             Pr(>Chi)
## NULL                       947     955.38
## Distance  1    137.8       946     817.58 < 0.00000000000000022 ***
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

We get more useful information from the coefficient `summary` of the model. This gives the intercept and slope of the model **on the scale of the linear predictor** (see the figure above).

```
summary(nflMod)
```

```
##
## Call:
## glm(formula = Success ~ Distance, family = binomial, data = NFL)
##
## Deviance Residuals:
##     Min       1Q   Median       3Q      Max
## -2.6681   0.2704   0.4067   0.7094   1.4708
##
## Coefficients:
##             Estimate Std. Error z value            Pr(>|z|)
## (Intercept)  5.68958    0.45021   12.64 <0.0000000000000002 ***
## Distance    -0.13118    0.01263  -10.38 <0.0000000000000002 ***
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## (Dispersion parameter for binomial family taken to be 1)
##
##     Null deviance: 955.38  on 947  degrees of freedom
## Residual deviance: 817.58  on 946  degrees of freedom
## AIC: 821.58
##
## Number of Fisher Scoring iterations: 5
```

We could report this something like this:

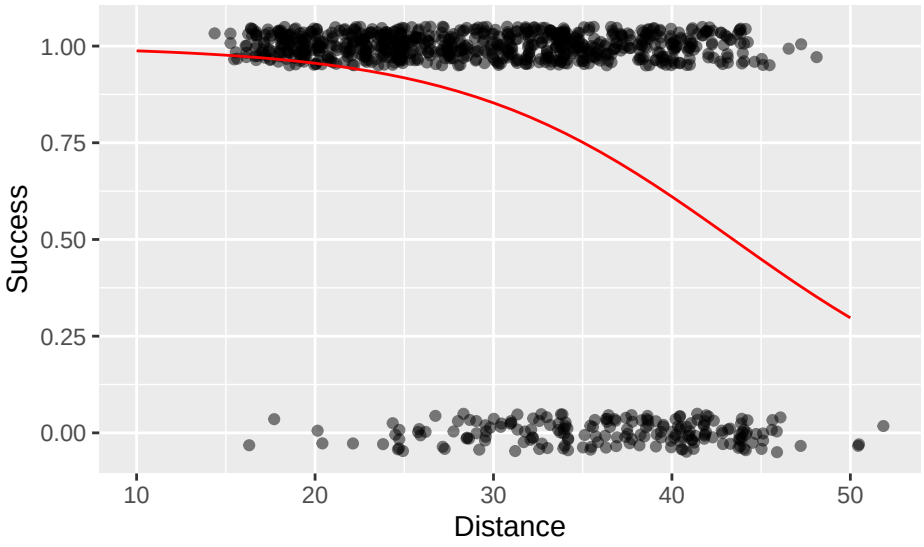
The binomial GLM showed that distance was significantly associated with the probability of goal success (GLM: Null Deviance = 955.38, Residual Deviance = 817.58, d.f. = 1 and 946 p <0.001). The slope and intercept of the relationship is -0.131 and 5.690 respectively on the logit scale. The equation of the best fit line was therefore logit(nOffspring) = 5.690 - 0.131×distance (see Figure XXX)

The equation of the model if you want to express it on the natural scale works out to be:

$y = \frac{1}{1+exp(-(\beta_0+\beta_1 x))}$, or $probability = \frac{1}{1+exp(-(5.690-0.131 \times distance))}$.

Let's make sure that works by creating a set of predicted data from this equation and plotting it onto the graph:

```
d1 <- data.frame(x = 10:50) %>%
  mutate(y = 1 / (1 + exp(-(5.690 - 0.131 * x))))
A +
  geom_line(data = d1, aes(x, y), colour = "red")
```



Rather than using the equation, there is an easier way by using the **R**'s `predict` function. We will use `predict` to get the predicted probability of success across the range of distances that we provide as a new data frame that we are here calling `newDat`. We will also predict the 95% Confidence Interval (CI) for these estimates. We will use these data to add the CI ribbon and line to the plot.

The `predict` function returns an object that includes the `fit` and the `se.fit` which are the predicted value of the regression and the standard error of that predicted value respectively. Thus, if the output of the `predict` function is stored as `pv` we can address those parts as `pv$fit` and `pv$se.fit`, and we can save these values alongside the data in `newDat`.

> Remember that the model, and the predicted values from it, are expressed on the scale of the *linear predictor* (i.e. the transformed data scale). It is usually desirable to "backtransform" to the natural scale before plotting. See below.

First we obtain the predictions and CI on the scale of the linear predictor (logit scale). We calculate 95% CI from the standard errors simply by multiplying by 1.96.

```
# Dataset to predict FROM
newDat <- data.frame(Distance = 14:52)
```

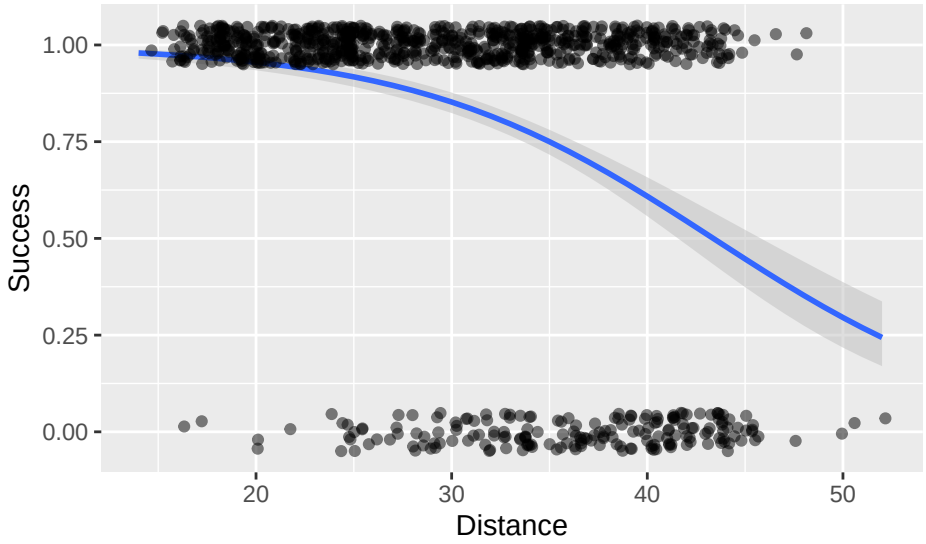
```
# Get predictions from the model
pv <- predict(nflMod, newdata = newDat, se.fit = TRUE)

# Add those predictions to newDat
newDat <- newDat %>%
  mutate(Success_LP = pv$fit) %>%
  mutate(lowerCI_LP = pv$fit - 1.96 * pv$se.fit) %>%
  mutate(upperCI_LP = pv$fit + 1.96 * pv$se.fit)
```

Now we can first obtain the inverse link from the model object `family(nflMod)$linkinv`, and use that to backtransform the data onto the natural scale ready for plotting.

```
# Get the inverse link function
inverseFunction <- family(nflMod)$linkinv

# transform predicted data to the natural scale
newDat <- newDat %>%
  mutate(
    Success = inverseFunction(Success_LP),
    ymin = inverseFunction(lowerCI_LP),
    ymax = inverseFunction(upperCI_LP)
  )
```

Now we can finally plot the model predictions and the 95% confidence intervals for them.

```
# The plot and ribbon
(A <- ggplot(NFL, aes(x = Distance, y = Success)) +
  geom_ribbon(
    data = newDat, aes(x = Distance, ymin = ymin, ymax = ymax),
    fill = "grey75", alpha = 0.5
  ) +
  geom_smooth(data = newDat, stat = "identity") +
  geom_jitter(height = 0.05, alpha = 0.5))
```



So, at what distance does the probability of success fall to just 50%? You could read this directly from the plot as "approximately 44m". Alternatively, you could obtain the value from the `newDat` dataset you created above with predictions from the model, by filtering it. This confirms the probability of success reaches 0.5 somewhere between 43-44m.

```
newDat %>%
  filter(Success < 0.55) %>%
  filter(Success > 0.45) %>%
  select(Distance, Success)
```

```
##   Distance   Success
## 1       42 0.5449184
## 2       43 0.5122432
## 3       44 0.4794630
```

20.3 Example: Sex ratio in turtles

In this example we will look at sex ratio of hawksbill turtles (*Eretmochelys imbricata*)[2]. The data are counts of males and females in clutches of eggs incubated at different temperatures.

The sex ratio in the species varies with temperature during incubation. We are interested in what the "tipping point" temperature is between male-female biased ratios.

[2]data from: Godfrey et al. (1999) Can. J. Zool. 77: 1465–1473

```
hawksbill <- read.csv("CourseData/hawksbill.csv")
```

This is a small dataset, you can look at the whole thing:

```
hawksbill
```

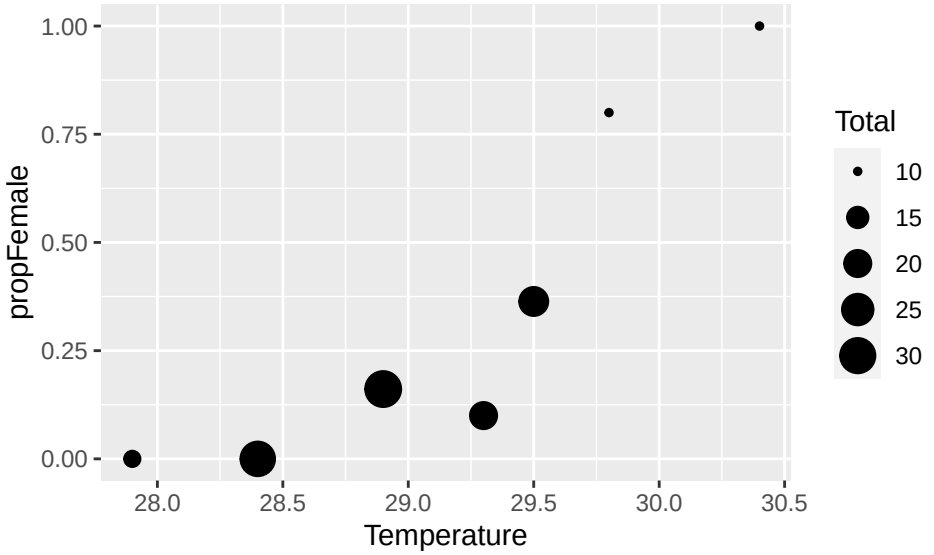
```
##   Temperature Total Nmale Nfemale
## 1        27.9    12    12       0
## 2        28.4    29    29       0
## 3        28.9    31    26       5
## 4        29.3    20    18       2
## 5        29.5    22    14       8
## 6        29.8    10     2       8
## 7        30.4    10     0      10
```

We are interested in sex ratio which we can calculate as the proportion of the population that is female (i.e. number of females divided by total number).

```
hawksbill <- hawksbill %>%
  mutate(propFemale = Nfemale / (Nmale + Nfemale))
```

Let's plot that data. We can use the trick of telling R to plot the points different sizes depending on the sample size (using the `size =` argument):

```
(A <- ggplot(hawksbill, aes(
  x = Temperature, y = propFemale,
  size = Total
)) +
  geom_point())
```



You can see that the proportion of females increases with temperature. Let's fit a model to these data to better understand them. We could use the `propFemale` data as the response variable, but there is a big problem with that: we would be giving equal weight to each of the data points, even though the sample size for each one ranges from 10 to 31 This is not good because we would have much more faith in the very large sample sizes than the small ones.

Instead we can bind the data into a two column matrix using `cbind`. The first column is our "success" and the second column is our "failure". If we put `females` in the first column the model will predict "probability of being female", which is what we want. This two-column approach provides the model with the sample size which it can use to weight the regression appropriately.

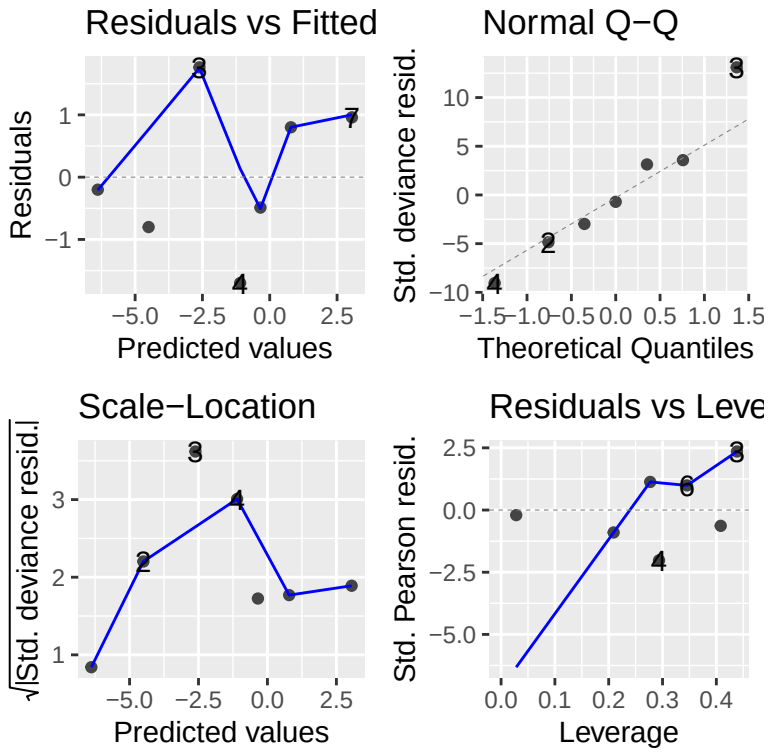
```
y <- cbind(hawksbill$Nfemale, hawksbill$Nmale)
```

Now lets fit the model.

```
modA <- glm(y ~ Temperature, data = hawksbill, family = binomial)
```

As ever, we should take a quick look at the model diagnostic plots - these look OK.

```
library(ggfortify)
autoplot(modA)
```



Now we can look at the anova table. This will tell us what we already know - there is a strong effect of temperature on sex ratio.

```
anova(modA, test = "Chi")
```

```
## Analysis of Deviance Table
##
## Model: binomial, link: logit
##
## Response: y
##
## Terms added sequentially (first to last)
##
##
##             Df Deviance Resid. Df Resid. Dev             Pr(>Chi)
## NULL                            6     70.353
## Temperature  1   61.869         5      8.484 0.000000000000003671 ***
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

Now for the coefficients:

```
summary(modA)
```

```
##
## Call:
## glm(formula = y ~ Temperature, family = binomial, data = hawksbill)
##
## Deviance Residuals:
##       1        2        3        4        5        6        7
## -0.2010  -0.8013   1.7632  -1.6999  -0.4885   0.8010   0.9607
##
## Coefficients:
##              Estimate Std. Error z value    Pr(>|z|)
## (Intercept) -111.7200    22.4260  -4.982 0.000000630 ***
## Temperature    3.7754     0.7625   4.951 0.000000737 ***
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## (Dispersion parameter for binomial family taken to be 1)
##
##     Null deviance: 70.3528  on 6  degrees of freedom
## Residual deviance:  8.4841  on 5  degrees of freedom
## AIC: 24.184
##
## Number of Fisher Scoring iterations: 5
```

This table gives us the coefficients for the formula of our relationship. We could use these to produce a formula of the form $y = \frac{1}{1+exp(-(\beta_0+\beta_1 x))}$.

It is perhaps more useful to plot the model fit onto the plot. First we need to create a data frame to predict from:

```
newDat <- data.frame(Temperature = seq(27.9, 30.4, 0.1))
```

Now we can predict the values (and 95% CI) on the scale of the linear predictor (logit).

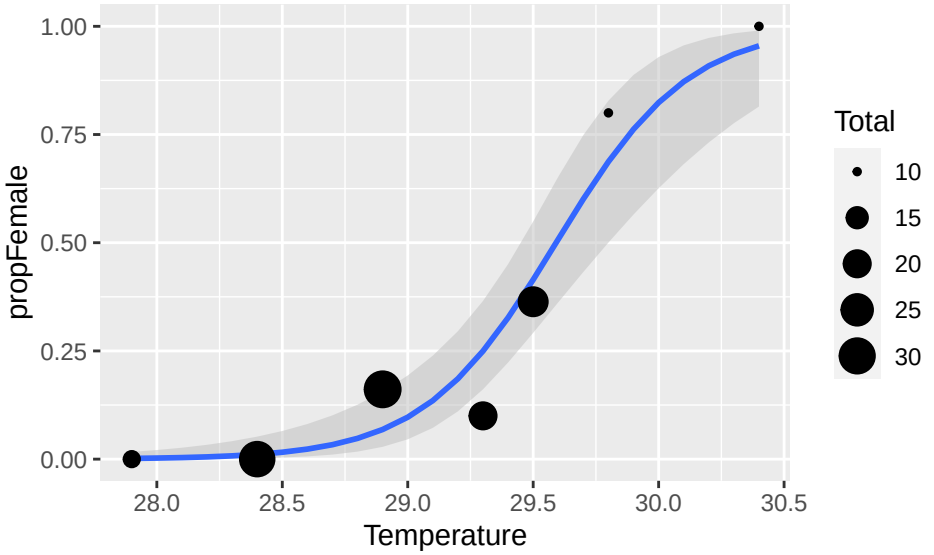
```
pv <- predict(modA, newDat, se.fit = TRUE)
newDat <- newDat %>%
  mutate(
    propFemale_LP = pv$fit,
    lowerCI_LP = pv$fit - 1.96 * pv$se.fit,
    upperCI_LP = pv$fit + 1.96 * pv$se.fit
  )
```

Now we can use the inverse link function to backtransform to the natural probability scale.

```r
# Get the inverse link function
inverseFunction <- family(modA)$linkinv

# transform predicted data to the natural scale
newDat <- newDat %>%
  mutate(
    propFemale = inverseFunction(propFemale_LP),
    ymin = inverseFunction(lowerCI_LP),
    ymax = inverseFunction(upperCI_LP)
  )
```

We can add these values to the plot like this:

```r
# The plot and ribbon
(A <- ggplot(hawksbill, aes(
  x = Temperature, y = propFemale,
  size = Total
)) +
  geom_ribbon(
    data = newDat, aes(x = Temperature, ymin = ymin, ymax = ymax),
    fill = "grey75", alpha = 0.5, inherit.aes = FALSE
  ) +
  geom_smooth(
    data = newDat, aes(x = Temperature, y = propFemale),
    stat = "identity", inherit.aes = FALSE
  ) +
  geom_point())
```



Can you give read off the graph the approximate estimated temperature at which sex ratio is 50:50?

> Try refitting the model using simply the proportion female (`propFemale`) data rather than the two-column (`cbind`) approach. Then try writing up the result in the same way as shown for the NFL field goals example.

20.4 Example: Smoking

As I mentioned above binomial regression can be applied to anything where there data can be classified into two groups. I'll illustrate that now with an example about smoking.

The data set is very small and looks like this:

	Student smokes	Student does not smoke
Parent(s) smoke	816	3203
No parents smoke	188	1168

The dataset is the number of students smoking and not smoking grouped according to whether their parents smoke. These data are binomial/proportion data because the values in the cells of the table are constrained by the overall total and students must fall into one of the categories. We can use these data to calculate the probability of the student being a smoker.

Before fitting a GLM lets just work out these probabilities by hand. What is the probability that a child of smoking parents is a smoker themselves? This is simply 816/(816+3203) = 0.2030. (i.e. it is the number of smokers divided by the total number). Similarly, the probability that the child of non-smokers smokes is 188/(188+1168) = 0.1386.

But is this a significant difference? That is what we are trying to find out using a GLM.

To do this, we can turn these data into a two column matrix of success (yes - smoker) and failure (no - non-smoker).

```
y <- cbind("1_yes" = c(816, 188), "0_no" = c(3203, 1168))
y
```

```
##      1_yes 0_no
## [1,]   816 3203
## [2,]   188 1168
```

So we can see “success” on the left and “failure” on the right“. We now create a (tiny) `data.frame` for the parental status (smoker =”1_yes“, non-smoker =”0_no”).

```
smoke <- data.frame(parentSmoke = c("1_yes", "0_no"))
```

Now we can fit the model. Pause now and think about what the NULL hypothesis is here. It is that parental smoking does not have any influence on whether the child smokes, and that the probability that the student smokes is unrelated to parental smoking.

```
smokeMod <- glm(y ~ parentSmoke, data = smoke, family = binomial)
```

With such a small dataset, diagnostic plots will not tell us anything useful so there's no point in doing them for this case.

As usual, we first ask for the (Analysis of Deviance table using `anova`.

```
anova(smokeMod, test = "Chi")
```

```
## Analysis of Deviance Table
##
## Model: binomial, link: logit
##
## Response: y
##
## Terms added sequentially (first to last)
##
##
##             Df Deviance Resid. Df Resid. Dev      Pr(>Chi)
## NULL                              1     29.121
## parentSmoke  1   29.121         0      0.000 0.00000006801 ***
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

This tells us that the status of the parents (whether they smoke or not) is highly significant: it explains a lot of the pattern that we see in the data.

We can find out what this pattern is by examining the `summary` table.

```
summary(smokeMod)
```

```
##
## Call:
## glm(formula = y ~ parentSmoke, family = binomial, data = smoke)
```

```
##
## Deviance Residuals:
## [1]  0  0
##
## Coefficients:
##                  Estimate Std. Error z value             Pr(>|z|)
## (Intercept)      -1.82661    0.07858 -23.244 < 0.0000000000000002 ***
## parentSmoke1_yes  0.45918    0.08782   5.228          0.000000171 ***
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## (Dispersion parameter for binomial family taken to be 1)
##
##     Null deviance: 29.1206557459935773  on 1  degrees of freedom
## Residual deviance: -0.0000000000003717  on 0  degrees of freedom
## AIC: 19.242
##
## Number of Fisher Scoring iterations: 2
```

This shows us the estimates on the logit scale. We can use `predict` to get a sense for these predictions on the more intuitive probability scale. First we calculate the fitted values and 95% confidence intervals on the scale of the linear predictor (logit):

```
pv <- predict(smokeMod, smoke, se.fit = TRUE)
smoke <- smoke %>%
  mutate(
    prob_LP = pv$fit,
    lowerCI_LP = pv$fit - 1.96 * pv$se.fit,
    upperCI_LP = pv$fit + 1.96 * pv$se.fit
  )
```

Then we can backtransform these values to the probability scale using the inverse link function:

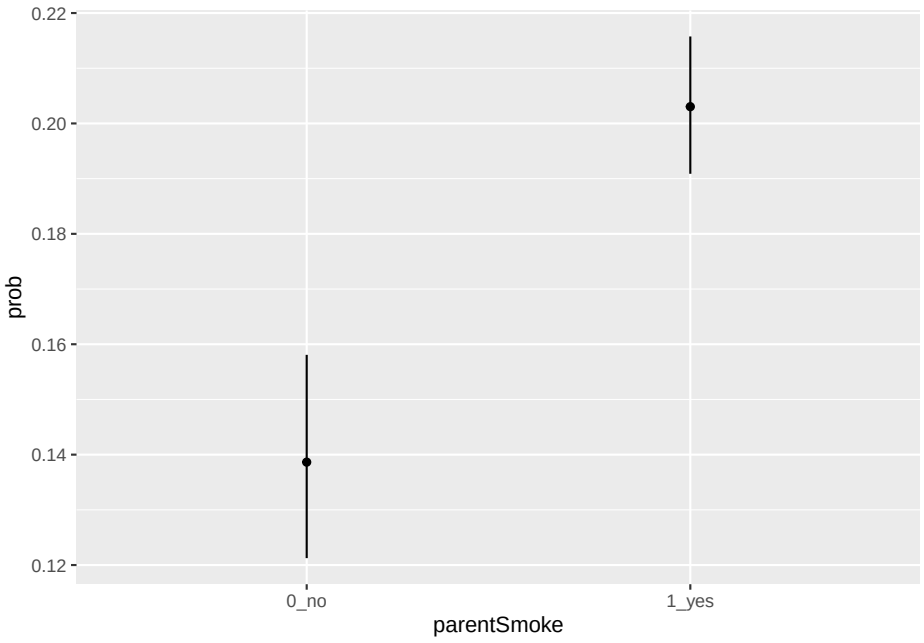
```
# Get the inverse link function
inverseFunction <- family(smokeMod)$linkinv

# transform predicted data to the natural scale
smoke <- smoke %>%
  mutate(
    prob = inverseFunction(prob_LP),
    ymin = inverseFunction(lowerCI_LP),
    ymax = inverseFunction(upperCI_LP)
  )
smoke
```

```
##   parentSmoke   prob_LP lowerCI_LP upperCI_LP      prob      ymin      ymax
```

```
## 1       1_yes -1.367429  -1.444287  -1.290570 0.2030356 0.1908823 0.2157563
## 2        0_no -1.826606  -1.980629  -1.672583 0.1386431 0.1212518 0.1580801
```

This table shows us that the probability of the students smoking is 0.2030 (95% CI = 0.191-0.216) for children of smokers, and 0.139 (95% CI = 0.121-0.158) for children of non-smokers. We can plot this using `ggplot` like this.

```
ggplot(smoke, aes(
  x = parentSmoke, y = prob, ymin = ymin,
  ymax = ymax
)) +
  geom_point() +
  geom_segment(aes(xend = parentSmoke, y = ymin, yend = ymax))
```



Chapter 21

Power analysis by simulation

This chapter will first focus on how we can answer questions like "*what sample size should I use in my experiment?*" and "*with this sample size, what difference could I detect?*"

As you learned in the chapters on t-tests and ANOVA, the detection of a significant difference between treatment groups (if there is one) depends on two things: (1) the *actual* difference between mean values for the groups (the "*signal*") and (2) the amount of variation there is in the groups (the "*noise*"). When there is a lot of noise it is hard to detect the signal.

In most cases we will already have some idea about what to expect when doing a study. Previous work on similar topics, or pilot studies, will have given us an idea of typical values for the response variable, and will give us a ballpark estimate of how much variation to expect. This information can be used to conduct a **power analysis by simulation**.

The basic idea of this approach is to simulate the experiment, by drawing random numbers from appropriate distributions, before actually carrying out the experiment.

21.1 Type I and II errors and statistical power

Before setting out to run an experiment or an observational study is is natural to wonder "*how much work do I really need to do here?*" In other words, "*what sample size do I need in order to address the hypothesis?*

Similar questions are also relevant **after** running an experiment. For example, imagine you have run an experiment that failed to find a significant effect of your treatment. There are two explanations for this finding (i) there really is no effect of your treatment; (ii) there **is** an effect of your treatment but you did not have enough power to detect it. So the question arising is: "what difference

could you have detected, based on the results you have?" The answer could be "*Based on my experiment I can see that if there* ***is*** *really a difference it must be smaller than X*" and/or "I would need to increase my sample size to X to detect a difference if the difference is *Y*"

These type of questions are closely related to the two types of error that one can make when testing hypotheses:

- A **Type I** error is the rejection of a true null hypothesis. For example, when there truly is no effect of an experimental treatment, but you detect one in error.
- A **Type II** error is the failure to reject a false null hypothesis. For example, when there truly is an effect of an experimental treatment, but you fail to detect it.[1]

The probability of making these errors depends on the **statistical power** of your study. Statistical power ranges between 0 and 1 and, for a simple hypothesis test, it is defined as **the probability that the test rejects the null hypothesis (H_0) when the alternative hypothesis (H_1) is true**. In other words it is the probability of NOT making a Type II error.

For example, a power of 0.80 means that you have an 80% chance of correctly detecting that H_1 is true and a 20% chance (1.0-0.8 = 0.2) of making a Type II error. As power decreases, the probability of making a Type II error increases.

21.2 What determines statistical power?

Statistical power is determined by several factors including the actual effect size, the natural variation in the effect size, the sample size in the study, and the (arbitrary) criterion you have chosen for significance:

1) The **magnitude of the actual effect** of interest. If the effect under investigation is very small then it will be harder to detect (i.e. the power to detect it will be small).
2) **Variation**. Variation in the data introduces noise into the statistical test. Where there is large amounts of natural variation it is harder to detect significant effects (i.e. the power to detect it will be reduced).
3) The **sample size**. Larger sample sizes reduce the amount of *sampling error* and therefore reduce the amount of "noise" in the data. Therefore large sample sizes increase power to detect a difference between groups.
4) Sampling error can be reduced by improving the precision of measurements. Therefore power can also be increased by improving measurement precision.
5) The **significance criterion**. We normally use p = 0.05, but this is arbitrary. We could increase the power of the statistical test by using a more relaxed criterion e.g. p = 0.10.

[1] To remember this, think of the story about "*the boy who cried wolf*". First the villagers believe the boy when there was no wolf (Type I error). Second, the villagers don't believe the boy but there IS a wolf (Type II error).

There are several ways to estimate statistical power, required sample sizes etc. In this chapter we will look at one of them – power analysis by simulation. This is best communicated using a simple example.

21.3 An example of calculating statistical power.

In an experiment, researchers would like to test a hypothesis that a high protein diet increases the size of adult insects from a pest species. Previous work has shown that the average size on a normal diet is 12mm with a standard deviation of 4mm.

> Remember to load the `dplyr`, `magrittr` and `ggplot` packages, and to set your working directory correctly.

We can simulate a distribution of measurements from this distribution using the `rnorm` function. This draws `n` numbers from a theoretical normal distribution with a particular `mean` and standard deviation (`sd`). For example:

```
(control <- rnorm(n = 10, mean = 12, sd = 3))
```

```
##  [1] 16.11288 10.30591 13.08939 13.89859 13.21280 11.68163 16.53457 11.71602
##  [9] 18.05527 11.81186
```

Because this is a random process your results will be different (and they will be different each time you run the code).

Suppose that other studies on a different species has shown that a high protein diet leads to a 20% increase in size. This means that we expect our treatment group to have a body length of $12 \times 1.2 = 14.4$mm. If we assume the standard deviation will be the same, we can simulate a distribution for a high protein treatment group in the same way as for the control group:

```
(treatment <- rnorm(n = 10, mean = 14.4, sd = 3))
```

```
##  [1] 18.16416 19.51226 17.04097 12.65537 13.89243 17.59434 16.55513 14.51473
##  [9] 11.25101 15.03694
```

Now we have two simulated samples, and we can conduct a t-test on those samples and store the result:

```
res <- t.test(control, treatment)
```

We can print this result to the screen like this:

```
res
```

```
##
##  Welch Two Sample t-test
##
## data:  control and treatment
## t = -1.7337, df = 17.976, p-value = 0.1001
## alternative hypothesis: true difference in means is not equal to 0
## 95 percent confidence interval:
##  -4.3793101  0.4196255
## sample estimates:
## mean of x mean of y
##  13.64189  15.62173
```

Or just get the p-value like this:

```
res$p.value
```

```
## [1] 0.1000936
```

21.3.1 The simulation

The idea with power analysis by simulation is to repeat this procedure many times (at least 1000) to estimate the probability that you get it right (that you detect a significant difference between groups when you know there is a difference[2]).

You have come across this sort of simulation approach before when we talked about randomisation tests. As before we will use the `replicate` function to repeatedly do t-tests on randomly generated data with the characteristics you select (i.e. sample size, mean, and standard deviation).

We first set up our simulation by defining the mean, standard deviation, and sample size of our simulated experiment.

```
controlMean <- 12
treatmentMean <- 14.4
sdValue <- 3
sampleSize <- 10
```

Now we can "wrap" a `t.test` on simulated control and treatment data sets within a `replicate` function like this. Take some time to study this part of the script - it is important that you understand what it is doing. In this case the replicate command is telling R to repeat the t-test 5 times.

[2]You know there is a difference because you have set this difference!

```
replicate(
  5,
  t.test(
    rnorm(sampleSize, controlMean, sdValue),
    rnorm(sampleSize, treatmentMean, sdValue)
  )$p.value
)
```

```
## [1] 0.0506748916 0.0004174904 0.0303818981 0.4768898365 0.0296402507
```

Let's repeat the t-test 1000 times so we can get a good idea of the number of times that the test correctly detects that there is a difference between the two groups.

I don't want to print 1000 p-values to the screen so I will collect them in a vector called `powerResults`.

```
powerResults <- replicate(
  1000,
  t.test(
    rnorm(sampleSize, controlMean, sdValue),
    rnorm(sampleSize, treatmentMean, sdValue)
  )$p.value
)
```

I can now ask how many of the p-values stored in `powerResults` were less than 0.05.

```
sum(powerResults < 0.05)
```

```
## [1] 393
```

So in this case, 393 of the 1000 tests were correct. The statistical power can be calculated by turning this into a percentage - i.e. Power = 39.3%.

We can ask what sample size do we need to get a better power, e.g. 90%, by increasing sample size incrementally. For example, here I increase sample size to 15:

```
controlMean <- 12
treatmentMean <- 14.4
sdValue <- 3
sampleSize <- 15 # Increased

powerResults <- replicate(
```

```
  1000,
  t.test(
    rnorm(sampleSize, controlMean, sdValue),
    rnorm(sampleSize, treatmentMean, sdValue)
  )$p.value
)

(sum(powerResults < 0.05) / 1000) * 100
```

```
## [1] 54.9
```

The power has increased to 54.9%.

21.3.2 Some questions for you to address:

1) What sample size would give you 80% power?

2) What power would you have if the variation was larger (e.g. sd = 4)?

3) What power would you have if the difference between groups was only 10% instead of 20%?

21.4 Summary

Simulation can be a powerful tool to help design and understand the results of hypothesis-based studies. The example above focuses on data that are normally distributed and where the test involved is a t-test. However, the same principles apply for other types of data. One can adapt the approach to use different distributions e.g. `rpois` for Poisson or `rbinom` for binomial. One can also alter the tests being done. This case study used a `t.test` but one could alternatively simulate the results of other ordinary linear models with `lm` or GLMs with `glm`.

21.5 Extending the simulation (optional, advanced)

This section is for illustration only. It is intended to show the utility of using R to quickly address experimental design questions but it goes beyond what you are expected to learn.I hope you find it interesting nevertheless. To this you will need to add the `purrr` library (using `install.packages("purrr")`). This package includes a useful function, `map_dbl`, which allows you to repeatedly apply functions over many different input values. I use the double colon (`::`) notation like this `purrr::map_dbl` to use this function without the need to load the whole package.

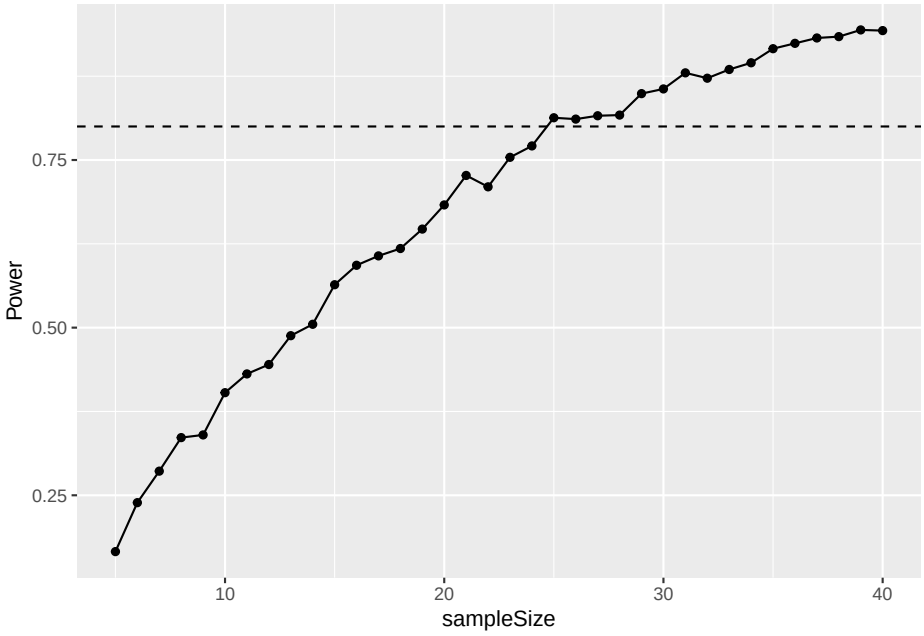
One can extend the simulation to cover a range of sample sizes (or differences between treatments, or standard deviations etc.) using by turning the calculations of the t-test into a function, and then applying that function over a range of values.

I illustrate this below by varying sample size between 5 and 40.

```
# Set up a data frame for the simulation results
simulData <- data.frame(sampleSize = 5:40)

# Set basic values
controlMean <- 12
treatmentMean <- 14.4
sdValue <- 3

# Function to do the t-test
pwr <- function(n) {
  sum(replicate(
    1000,
    t.test(
      rnorm(n, controlMean, sdValue),
      rnorm(n, treatmentMean, sdValue)
    )$p.value
  ) < 0.05) / 1000
}

# map_dbl applying the function for every value of
# simulData$sampleSize
simulData$Power <- purrr::map_dbl(simulData$sampleSize, pwr)
```

These results can then be graphed with `ggplot`.

```r
# Plot the output
ggplot(simulData, aes(x = sampleSize, y = Power)) +
  geom_point() +
  geom_line() +
  geom_hline(yintercept = 0.8, linetype = "dashed")
```



21.6 Exercise 1: Snails on the move

Your supervisor has collected pilot study data on the distance travelled by a particular snail species during one day. In this study the mean distance travelled was 7.4m and there was a standard deviation of 2.76m. There are two colour morphs of the snails: One is bright and colourful with a striking black spiral while the other is drab and kinda boring-looking. Your supervisor focused on the colourful snails, but assume that standard deviation is the same for both morphs.

For your masters project you hypothesise that the boring snail will cover more distance because it is less scared of visual predators and willing to expose itself to more risk while foraging. You don't have a good feel for the difference, but you decide that a 25% difference between the morphs would be biologically interesting.

Simulate a t-test-based analysis in R to figure out what sample size would result in 80% power.

21.7 Exercise 2: Mouse lemur strength

Previous work on grey mouse lemurs (*Microcebus murinus*) has found that they are quite strong. They can lift and hold 10 times their own body weight![3]

This work was done on prime-age animals. Researchers believe that older individuals will have experienced physiological senescence and that their strength will have deteriorated. The body weight of these lemurs is about 60grams. The prime-age animals could lift and hold 600grams with a standard deviation of 145grams.

A research institute has agreed to you carrying out this study for your masters project. They have 25 young age lemurs, but only have 8 old aged animals. You can assume the standard deviation is the same in both age classes.

What difference in strength could you reliably detect (with power >80%) with these sample sizes?

[3]Thomas, P., Pouydebat, E., Brazidec, M., Aujard, F., & Herrel, A. (2015). Determinants of pull strength in captive grey mouse lemurs Journal of Zoology DOI: 10.1111/jzo.12292

Part IV

Exam

Chapter 22

An example of a past Exam (2020)

This is a previous year's exam!

I am leaving this here so you can see what the final exam will look like.

This exam includes four questions that test different aspects of your learning during this course: data wrangling, data visualisation and statistics. Each question is broken down into a number sub-questions.
Your work should be handed in as a single PDF. The number of each question should be clearly indicated, and the answer to each question should start on a new page.
For each question you must provide the R code you used to answer the question. The code should include comments to explain what you are doing. The code should be provided as text using a fixed-width font such as `Courier`. The rest of your answers should be in another font (e.g. Times New Roman, Cambria). Please use *text* rather than a screenshot of your code.
You can use the Microsoft Word template provided alongside these questions (here) as guidance.

- Plots and tables should have appropriate captions.
- Plots should be produced using `ggplot`.
- Remember that you can make "panels" of plots (e.g. Fig 1A, B)
- Axis labels are important. Sometimes you may want to edit them to be different from a data column name.
- Reporting of any statistics should be appropriate to the type of analysis you have done. There are examples in the course materials.
- Reporting of methods and results should be written in the style of a scientific paper (again, there are examples in the course materials).

If you don't understand any aspects of the questions, please ask for help!
Hand in deadline is 8th January 2021 at 23:59 CET
You MUST submit your work via Blackboard (not email!)

---

1) Amazonian fires (10 points)

The dataset `amazon.csv` is a record of fire occurrence in the Amazon rainforest. The data set has columns for year, the state where the fire occurred, the month, and the number of fires reported. The month is recorded in Portuguese, so you will probably want to either create a "look up table" and use `left_join` to convert the month to a number, or use the `dplyr` function `recode`. There are many states and it might be useful to group them. For example the legal Amazon includes the states of Acre, Amapá, Pará, Amazonas, Rondonia, Roraima, Mato Grosso, Tocantins, and Maranhão.

a) Produce a graph showing the the total number of fires per year through time as points joined by lines.

b) Produce a table showing the minimum, maximum, mean and median number of fires per month in the legally-defined Amazon.

c) Produce a graph using box plots to show the distribution of the number of fires per month in the Amazon (i.e. month on x-axis, number of fires on y-axis).

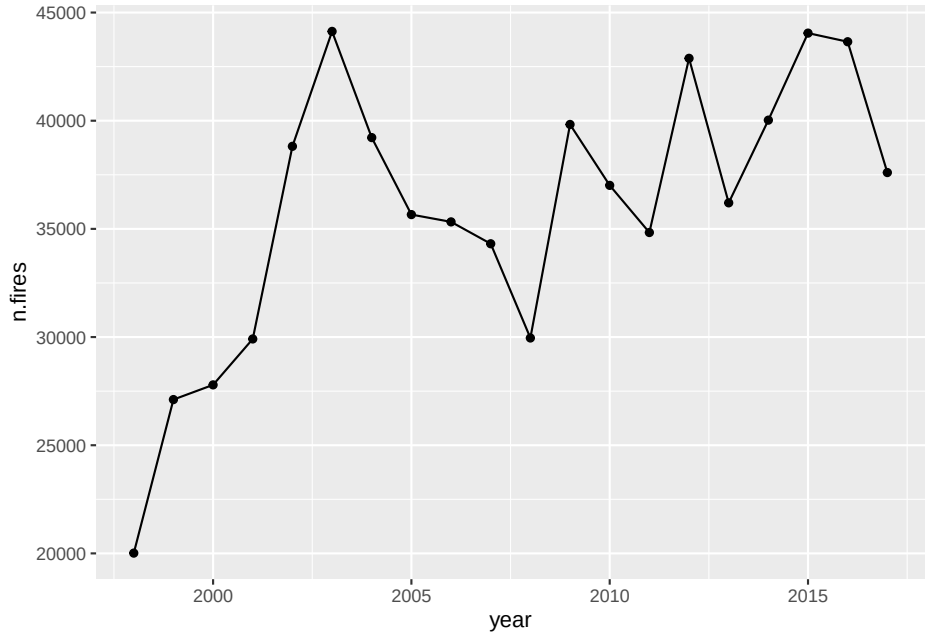
---

a) Produce a graph showing the the total number of fires per year through time as points joined by lines.

```
# Import data
amazon <- read.csv("CourseData/amazon.csv")
amazon <- read.csv("CourseData/amazon.csv") %>%
  mutate(number = ifelse(number >= 1000,
    1 + (number - 1000) / 1000, number
  ))

# A) MAKE GRAPH OF TOTAL FIRES PER YEAR.

(amazon_YearlyFires <- amazon %>%
  group_by(year) %>%
  summarise(n.fires = sum(number)))
```

```
## # A tibble: 20 x 2
##     year n.fires
##    <int>   <dbl>
##  1  1998  20014.
##  2  1999  27111.
##  3  2000  27790.
##  4  2001  29912.
##  5  2002  38816.
##  6  2003  44128.
##  7  2004  39220.
##  8  2005  35661.
##  9  2006  35324.
## 10  2007  34312.
## 11  2008  29957.
## 12  2009  39827.
## 13  2010  37011.
## 14  2011  34833.
## 15  2012  42884.
## 16  2013  36203.
## 17  2014  40026.
## 18  2015  44049.
## 19  2016  43651.
## 20  2017  37604.
```

```
(A <- ggplot(amazon_YearlyFires, aes(x = year, y = n.fires)) +
  geom_point() +
  geom_line())
```



You may have slightly different values if you used old data that had some fractional numbers (e.g. 1.543 is not "1 point 5" fires, but one thousand five hundred!). Well done those of you that spotted that issue. Don't worry if you didn't - you don't lose points!

Some of you may also have made plots that included ONLY data from the "legal amazon", by filtering to those states only. Again, that is no problem if you did that.

b) Produce a table showing the minimum, maximum, mean and median number of fires per month in the legally-defined Amazon.

```
# First filter to only include the "legally defined amazon"
# Check that all are included in the result (you might need to
# check and fix "foreign" letters - a typical data cleaning task!)
legalAmazonStates <- c(
  "Acre", "Amapa", "Pará", "Amazonas", "Rondonia",
  "Roraima", "Mato Grosso", "Tocantins", "Maranhao"
)

legalAmazon <- amazon %>%
  filter(state %in% legalAmazonStates)
```

```r
# Next make a "look up" table to convert month name
# to a numeric value 1-12
(monthLookup <- amazon %>%
  select(month) %>%
  unique() %>%
  mutate(month_numeric = 1:12))
```

```
##         month month_numeric
## 1     Janeiro             1
## 21  Fevereiro             2
## 41      Março             3
## 61      Abril             4
## 81       Maio             5
## 101     Junho             6
## 121     Julho             7
## 141    Agosto             8
## 161  Setembro             9
## 181   Outubro            10
## 201  Novembro            11
## 221  Dezembro            12
```

```r
# Then use left_join to add the numeric month to the data
# then group_by month and year, and summarise to get total
# number (i.e. sum for each month in each year for whole amazon)
# Then group_by month again and calculate the min/mean/max etc.
table1 <- legalAmazon %>%
  left_join(monthLookup) %>%
  group_by(month_numeric, year) %>%
  summarise(sumFires = sum(number)) %>%
  # total fires per month, per year
  group_by(month_numeric) %>%
  # group by month to get min/max/median per month
  summarise(
    minimum = min(sumFires), mean = mean(sumFires),
    median = median(sumFires), maximum = max(sumFires)
  )

table1
```

```
## # A tibble: 12 x 5
##    month_numeric minimum  mean median maximum
##            <int>   <dbl> <dbl>  <dbl>   <dbl>
##  1             1       0 1382.  1370.    3257
##  2             2       0  886.   801     1963
##  3             3       0  886.   857     1612
##  4             4       0  811.   792     1566
##  5             5       0  980.  1028.    1938
```

```
##  6             6   333.  1492.  1526.   2670
##  7             7   772.  2025.  2142.   3422.
##  8             8    46.2 1073.   978.   3337.
##  9             9   132.   864.   919.   1695.
## 10            10   594.  2064.  2140.   3113.
## 11            11   634.  1889.  2014.   3619.
## 12            12   260.  1251.  1214.   2861.
```

This question was HARD. It could also have been interpreted in a couple of different ways. For example, it could be interpreted as asking for the mean/median/max (etc.) by state. What I actually meant was to find the TOTAL across ALL of the legal Amazon for each month and each year (20 years * 12 months = 240 values), and then produce a table summarising the mean, min, max etc. of these values.

If you calculate the statistics "by state" as well as by year and month you end up with a different table, like this:

```
table1 <- legalAmazon %>%
  left_join(monthLookup) %>%
  group_by(month_numeric) %>%
  summarise(
    minimum = min(number), mean = mean(number),
    median = median(number), maximum = max(number)
  )

table1
```

```
## # A tibble: 12 x 5
##    month_numeric minimum  mean median maximum
##            <int>   <dbl> <dbl>  <dbl>   <dbl>
##  1             1       0 138.   44.5      960
##  2             2       0  88.6  26        871
##  3             3       0  88.6  19        820
##  4             4       0  81.1  17        947
##  5             5       0  98.0  12        942
##  6             6       0 149.   36        979
##  7             7       0 202.   39        989
##  8             8       0 107.    5.27     960
##  9             9       1  86.4   6.27     998
## 10            10       1 206.   13.5      960
## 11            11       0 189.  105        973
## 12            12       0 125.   46        846
```

I have been lenient with the marking on this one!

c) Produce a graph using box plots to show the distribution of the number of fires per month in the Amazon (i.e. month on x-axis, number of fires on y-axis).

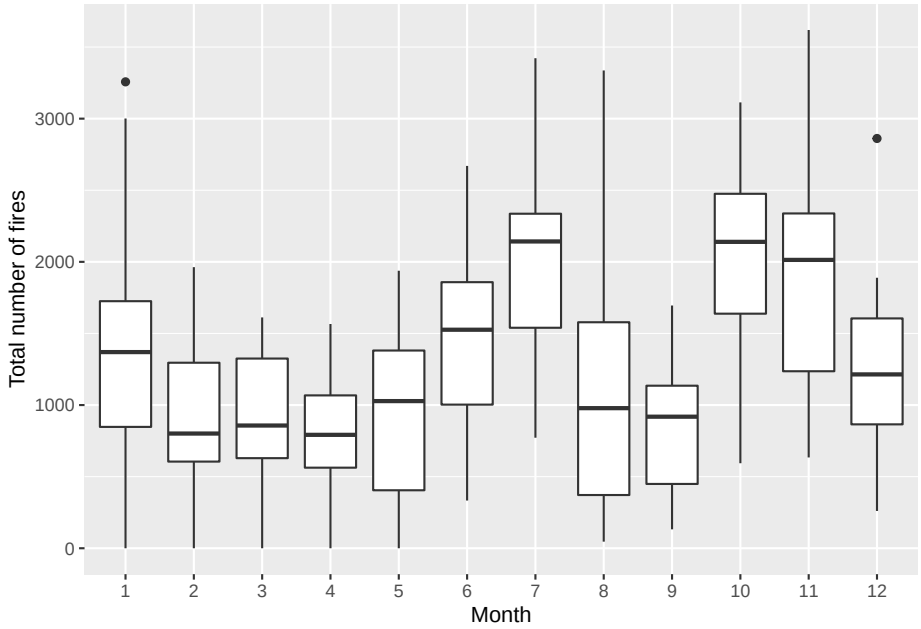
Here I am looking for a box plot of the number of fires per month in the Amazon (or legal Amazon). In other words, each row of data (each data point) should be the total number of fires across all of the states, for each month, in each year.

To check that you are on the right track you could look at the dimensions of the data frame you create - with 12 months per year and 20 years of data you can expect 240 rows of data (though perhaps not all years include all months of data). Nevertheless, it gives you a ballpark figure and if your dataset is not approximately this size, something has gone wrong!

```
# Use summarise to calculate the the total number of
# fires per month.
# In this case I use the legal amazon, but it is acceptable
# to look at the whole data set. You need to make sure that
# you are using the numeric month.

# Group by year and month to calculate the total number of
# fires in each year and month.

# Create the dataset by filtering to legal amazon, adding
# numeric month, grouping by year and month and finally
# summarising.
amazon2 <- amazon %>%
  filter(state %in% legalAmazonStates) %>%
  left_join(monthLookup) %>%
  group_by(year, month_numeric) %>%
  summarise(n_fires = sum(number))

# Check number of rows
nrow(amazon2)
```

```
## [1] 239
```

```
# Plot the data
ggplot(amazon2, aes(x = as.factor(month_numeric), y = n_fires)) +
  geom_boxplot() +
  xlab("Month") +
  ylab("Total number of fires")
```



Common mistakes - Understanding how `group_by` and `summarise` work is key to answering this question. To ensure that the months are displayed in the correct order you need to use the `numeric` month. Failure to do that will cause the plot to be presented in alphabetical order (i.e. with December falling before January!). However, box plots require that the input variable is a `factor` so you also need to convert back to a `factor` before plotting.

---

2) Coral bleaching (10 points)

Coral bleaching is when coral polyps expel the endosymbiotic algae that live within their tissues. Although the coral can survive bleaching events, their algae provide most of their energy, so the coral can eventually starve and die. It is thought that deeper corals (from the mesophotic zone) might be protected from bleaching events because the depth offers more stable conditions with fewer stressors. This is known as the "deep reef refugia hypothesis".

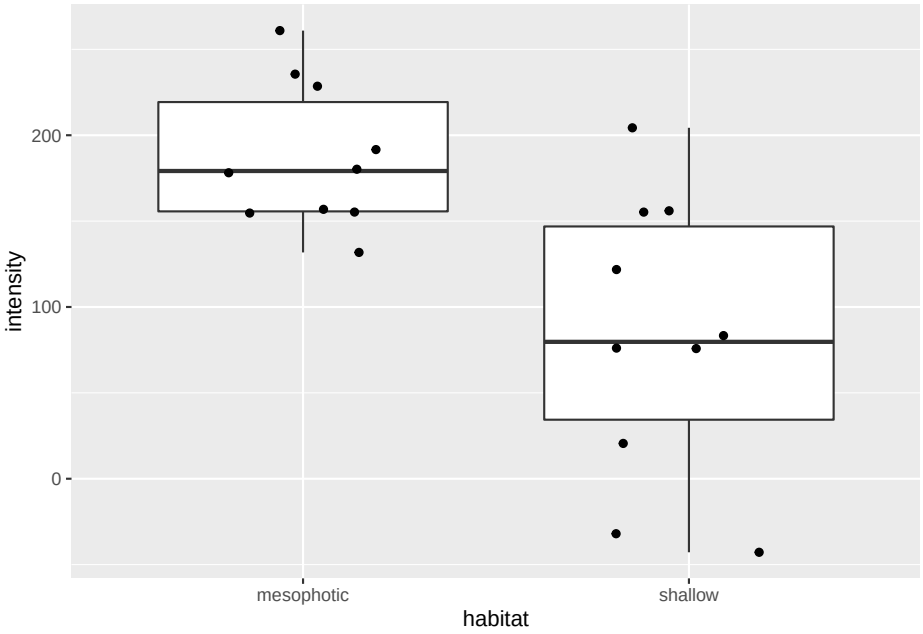
To test this idea, a transplant experiment was carried out on the coral *Agaricia lamarcki* at the island of Utila, Honduras. In the study, intact samples of the coral were moved from deep (mesophotic) reefs to the shallow reef and *vice versa*. They were left there for 8 months and then their colouration was measured to assess bleaching: lower colour intensity means more bleaching. The data are provided in `coral.csv`.

a) plot the data (e.g. with a box plot)

b) carry out a randomisation test to determine if there is a significant difference in the colour intensity in the two habitats. Write (i) a brief method description and (ii) a summary of the results.

a) plot the data (e.g. with a box plot)

```
coral <- read.csv("CourseData/coral.csv", stringsAsFactors = TRUE)

ggplot(coral, aes(x = habitat, y = intensity)) +
  geom_boxplot() +
  geom_jitter(width = 0.2)
```



b) carry out a randomisation test to determine if there is a significant difference in the colour intensity in the two habitats. Write (i) a brief method description and (ii) a summary of the results.

First we need to calculate the observed difference between the two habitats.

You can get the mean values like this (there are other ways):

```
coral %>%
  group_by(habitat) %>%
  summarise(meanIntensity = mean(intensity))
```

```
## # A tibble: 2 x 2
##   habitat    meanIntensity
##   <fct>              <dbl>
## 1 mesophotic         187.
## 2 shallow             81.8
```

…then subtract one value from the other "by hand".

You could also do the whole calculation using the pipes syntax like this:

```
(obsDiff <- coral %>%
  group_by(habitat) %>%
  summarise(meanIntensity = mean(intensity)) %>%
  pull(meanIntensity) %>%
  diff())
```

```
## [1] -105.5377
```

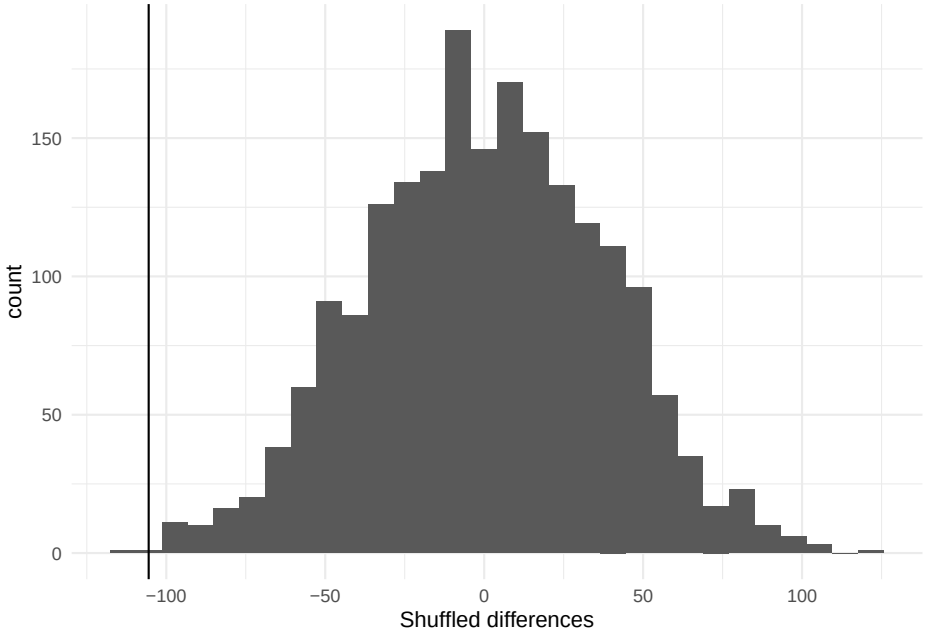
Next we ask "is the observed difference significantly larger than what we would expect by chance?".[1]

```
shuffledData <- data.frame(rep = 1:2000) %>%
  mutate(shuffledDiffs = replicate(
    2000,
    coral %>%
      mutate(habitat = sample(habitat)) %>%
      group_by(habitat) %>%
      summarise(meanIntensity = mean(intensity)) %>%
      pull(meanIntensity) %>%
      diff()
  ))
```

I can plot these data, and add a line to show where the observed value is

```
ggplot(shuffledData, aes(x = shuffledDiffs)) +
  geom_histogram() +
  theme_minimal() +
  xlab("Shuffled differences") +
  geom_vline(xintercept = obsDiff)
```

[1] I set options `options(dplyr.summarise.inform = FALSE)` to avoid the annoying messages.



Now I can calculate the significance by asking how many of she shuffled differences were more extreme than the observed value.

```
table(shuffledData$shuffledDiffs <= obsDiff)
```

```
##
## FALSE  TRUE
##  1999     1
```

In this case, NONE of the shuffled values are more extreme than the observed value, so the p-value can be reported as "p<0.001". You could write this up like this

To test whether the difference in intensity between the mesophotic and shallow coral habitats is statistically significant I did a 1000 replicate randomisation test with the null hypothesis being that there is no difference between the group means. The alternative hypothesis is that the mean for the mesophotic habitat is significantly lower than the mean for the shallow habitat. I compared the observed difference to this null distribution to calculate a p-value in a one-sided test.

The observed mean values of light intensity for the mesophotic and shallow corals were 187.4 and 81.8 respectively and the difference between them is therefore -105.6 (shallow - mesophytic). None of the 1000 null distribution replicates were as extreme my observed difference value. I conclude that the observed difference between the means of the two groups is statistically significant ($p < 0.001$)"

3) Power in a field experiment (10 points)

Scientists have developed a new eco-friendly fertiliser made from seaweed extract. You are planning a outdoor field experiment to test how effective it is at increasing crop yield in oilseed rape (*Brassica napus*). A standard industrial chemical fertiliser can increase yield by 30%, and you would like to know if the new seaweed fertiliser has a similar effect. You will grow the plant in a number of 4m x 4m field plots with two treatments: (i) control, with no additional fertiliser (ii) addition of seaweed fertiliser.

You have some preliminary data from an older study (`oilseed.csv`) which shows the normal crop yield (in kg/ha). Use this data to do your power analysis

a) Summarise the older study data to obtain mean and standard deviation.

b) Conduct a power analysis based on the pilot study data to estimate the number of samples required to carry out your experiment with 80% power. Describe the results of this power analysis.

c) Briefly describe a simple proposed experiment design to test the new seaweed fertiliser.

---

a) Summarise the older study data to obtain mean and standard deviation.

```
oilseed <- read.csv("CourseData/oilseed.csv",
  stringsAsFactors = TRUE
)

(controlData <- oilseed %>%
  summarise(meanVal = mean(yield), sdVal = sd(yield)))
```

```
##   meanVal    sdVal
## 1  958.25 277.7619
```

b) Conduct a power analysis based on the pilot study data to estimate the number of samples required to carry out your experiment with 80% power. Describe the results of this power analysis.

```
controlMean <- controlData$meanVal
treatmentMean <- controlData$meanVal * 1.3 # Increase of 30%
sdValue <- controlData$sdVal
sampleSize <- 10

powerResults <- replicate(
  1000,
```

```
  t.test(
    rnorm(sampleSize, controlMean, sdValue),
    rnorm(sampleSize, treatmentMean, sdValue)
  )$p.value
)

# How many times is the test successful?
sum(powerResults < 0.05)
```

```
## [1] 559
```

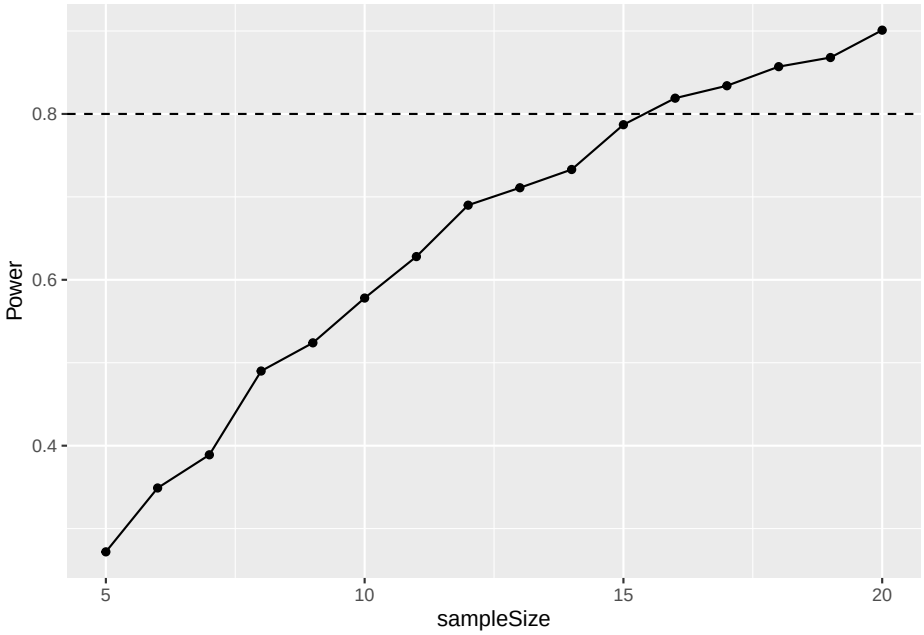
For 80% power, you want a sample size that results in 80% (i.e. 800 out of 1000 replicates) correctly detecting that there is a difference between control and treatment. Therefore you can increase `sampleSize` until you hit that magic number.

A more advanced approach is to write code to try a range of sample sizes, and collect the results so that they can be plotted in a graph showing the relationship between statistical power and samples size:

```
# Set up a data frame for the simulation results
simulData <- data.frame(sampleSize = 5:20)

# Set basic values
controlMean <- controlData$meanVal
treatmentMean <- controlData$meanVal * 1.3 # Increase of 30%
sdValue <- controlData$sdVal

# Function to do the t-test
pwr <- function(n) {
  sum(replicate(
    1000,
    t.test(
      rnorm(n, controlMean, sdValue),
      rnorm(n, treatmentMean, sdValue)
    )$p.value
  ) < 0.05) / 1000
}

# map_dbl applying the function for every value of
# simulData$sampleSize
simulData$Power <- purrr::map_dbl(simulData$sampleSize, pwr)
```

Now plot the results:

```r
# Plot the output
ggplot(simulData, aes(x = sampleSize, y = Power)) +
  geom_point() +
  geom_line() +
  geom_hline(yintercept = 0.8, linetype = "dashed")
```



From this, it looks like a power of 80% is achieved with a sample size of about 16.

Summing up: *My power analysis by simulation shows that a sample size of 16 per treatment group is required to detect a difference of 30% with 80% power.*

c) Briefly describe a simple proposed experiment design to test the new seaweed fertiliser.

You can write something like this:

Based on my power analysis, I would set up an experiment with 16 plots per treatment group. [you could also use more samples, and explain that you are adding extra as insurance in case of problems with some plots] I would try to ensure that the plots were as similar as possible to each other to avoid the effects of unmeasured differences between them. Then I would randomise the assignment of treatments (control vs. treated) to each plot. Ideally the plots would be spaced far apart to avoid pseudo-replication.

---

4) Biodiversity (20 points)

There is a well-known relationship between habitat area and biodiversity - the "species-area relationship". In a nutshell, the number of species tends to increase

as the area available increases. This relationship is clearly seen, for example, if we look at island biodiversity: larger islands support more species then small ones. One potential mechanism for this observed pattern is that larger islands tend to have more variety of different habitats, and therefore more niches available for species to occupy (more niches = more species).

The dataset `roundabouts.csv` shows the results of a study carried out in an urban environment to investigate these ideas. During the study, the number of beetle species (`nSpecies`) living on roundabouts and other "islands" of vegetation of different sizes (`area` in m^2) in a sea of concrete and tarmac was counted. Some of these islands had "complex" vegetation (e.g. trees, bushes, shrubs, ponds and rocks) while others were "simple" (only grass), indicated by the variable `habitatType`,

Use an appropriate statistical model to explore the relationship between area and species richness. Does this relationship differ depending on habitat complexity?

a) Plot the data to show the relationship between the the number of species and the area of the "island". Colour code the points by whether the habitat type.

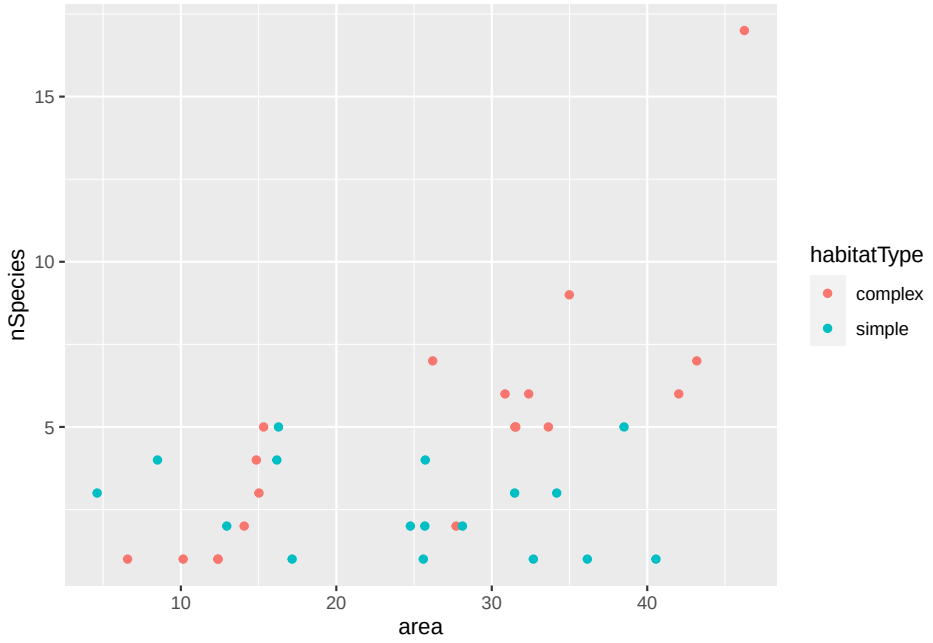
b) Fit a suitable statistical model to estimate the statistical relationship between area, habitat type, and species richness. Describe the method and then summarise the results produced by the model as if you were writing a report/thesis.

c) Produce a plot that shows (in addition to the raw data points) the fitted values produced by your model and the uncertainty in those estimates.

---

a) Plot the data to show the relationship between the the number of species and the area of the "island". Colour code the points by whether the habitat type.

```
roundabouts <- read.csv("CourseData/roundabouts.csv",
  stringsAsFactors = TRUE
)

ggplot(roundabouts, aes(
  x = area, y = nSpecies,
  colour = habitatType
)) +
  geom_point()
```

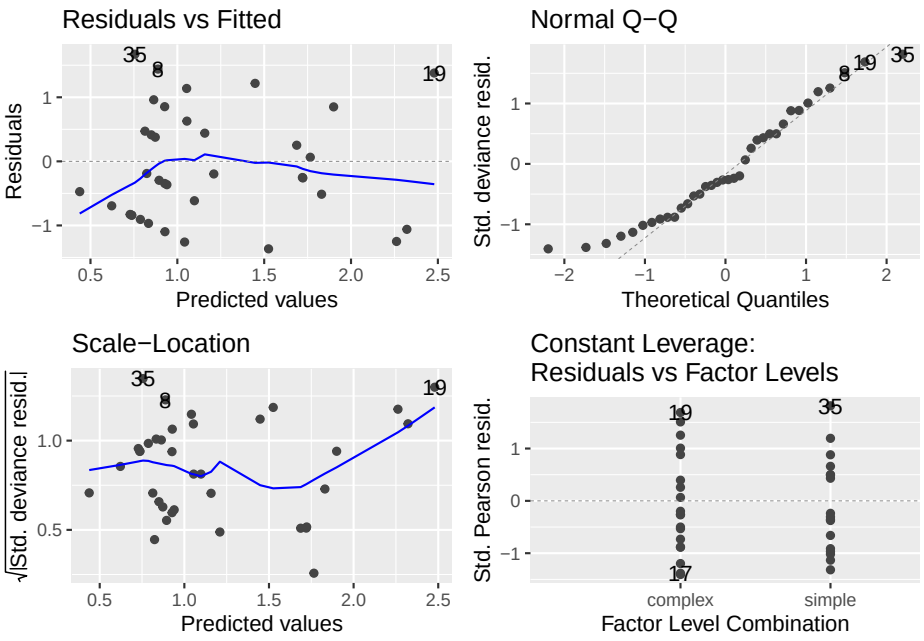


b) Fit a suitable statistical model to estimate the statistical relationship between area, habitat type, and species richness. Describe the method and then summarise the results produced by the model as if you were writing a report/thesis.

The key thing to remember here is that there are limits to counts of species. You can't count <0. Therefore, models that would allow the prediction of negative values may be problematic. Generalised Linear Models are designed for handling these kind of issues and, in this case, a Poisson model is most appropriate. This is an analogous situation to the example we looked at modelling the number of fox offspring in relation to mother's weight.

I could describe the methods like this: *I fitted a generalised linear model with Poisson errors to model how species richness is associated with survey area and habitat complexity. I included species richness as the response variable, and area and habitat complexity as explanatory variables. I used Poisson errors because these are count data and using an ordinary linear model could lead to estimates of negative species richness, and would not account for the fact that variance increases with the mean, which violates one of the assumptions of ordinary regression models. After fitting the model I examined the diagnostic plots of the model to check that the model was appropriate.*

```
mod1 <- glm(nSpecies ~ area * habitatType,
  family = "poisson",
  data = roundabouts
)
autoplot(mod1)
```




```
anova(mod1, test = "Chi")
```

```
## Analysis of Deviance Table
##
## Model: poisson, link: log
##
## Response: nSpecies
##
## Terms added sequentially (first to last)
##
##
##                   Df Deviance Resid. Df Resid. Dev    Pr(>Chi)
## NULL                                 35     72.607
## area               1   21.627        34     50.980 0.000003311 ***
## habitatType        1   10.945        33     40.035   0.0009386 ***
## area:habitatType   1   13.842        32     26.193   0.0001988 ***
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

```
summary(mod1)
```

```
##
## Call:
## glm(formula = nSpecies ~ area * habitatType, family = "poisson",
##     data = roundabouts)
##
```

```
## Deviance Residuals:
##     Min       1Q   Median       3Q      Max
## -1.3652  -0.8311  -0.2552   0.5103   1.6721
##
## Coefficients:
##                          Estimate Std. Error z value     Pr(>|z|)
## (Intercept)              0.099910   0.321923   0.310     0.756291
## area                     0.051446   0.009423   5.460 0.0000000477 ***
## habitatTypesimple        1.171997   0.488857   2.397     0.016511 *
## area:habitatTypesimple -0.064852   0.017255  -3.758     0.000171 ***
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## (Dispersion parameter for poisson family taken to be 1)
##
##     Null deviance: 72.607  on 35  degrees of freedom
## Residual deviance: 26.193  on 32  degrees of freedom
## AIC: 141.11
##
## Number of Fisher Scoring iterations: 4
```

I can describe the results like this:

The GLM results show that the effect of area and habitat type were both highly-significant (GLM, Area: null deviance= 72.707, residual deviance = 50.980, d.f. 1 and 34, $p < 0.001$; habitat type: null deviance= 72.707, residual deviance = 40.035, d.f. 1 and 33,$p < 0.001$)). There was also a significant interaction between area and habitat type (i.e. the effect of area depended upon the habitat) (interaction: null deviance= 72.707, residual deviance = 26.193, d.f. 1 and 32,$p < 0.001$)). The model showed that area was positively associated with species richness for complex habitats (slope = 0.05+/0.009) but that the slope (on the scale of the linear predictor of the GLM) was close to zero for simple habitats (slope = -0.01 [`0.051446 + (-0.064852)`]) (see Figure x)

c) Produce a plot that shows (in addition to the raw data points) the fitted values produced by your model and the uncertainty in those estimates.

Here I am asking you to add the predicted values (fitted value lines) from your model to the plot of the raw data. I am also asking you to show uncertainty. This is best done by adding a shaded ribbon.

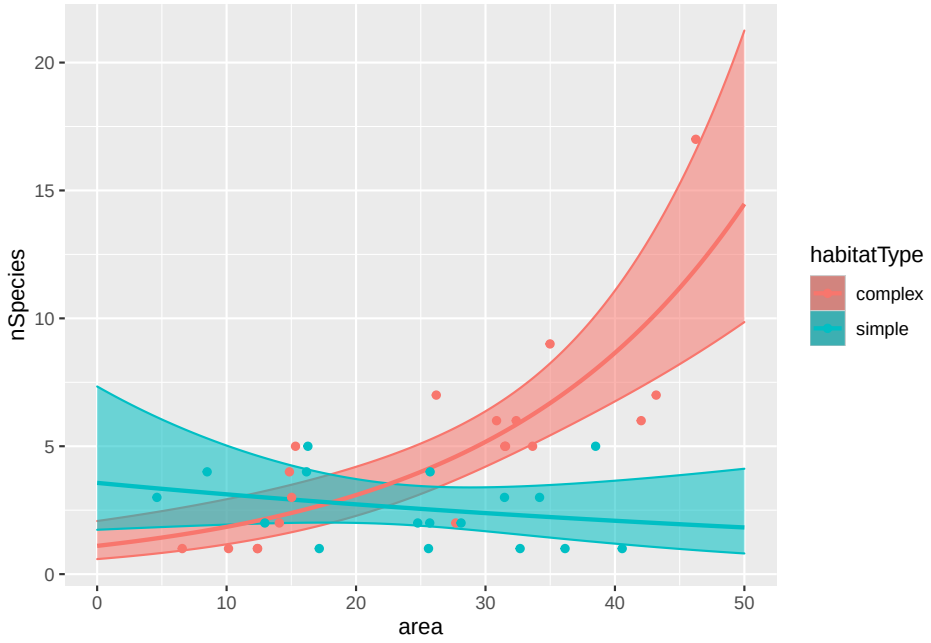
The main thing to remember here is that you need to back-transform from the scale of the linear predictor to the natural scale.

```
# Data set to predict from
newData <- expand.grid(
  area = seq(0, 50, 0.1),
  habitatType = c("complex", "simple")
)
```

```
# Predicted values (and SE)
pv <- predict(mod1, newData, se.fit = TRUE)

# Create new data for the predicted fit line
newData <- newData %>%
  mutate(nSpecies_LP = pv$fit) %>%
  mutate(lowerCI_LP = pv$fit - 1.96 * pv$se.fit) %>%
  mutate(upperCI_LP = pv$fit + 1.96 * pv$se.fit)

# Get the inverse link function
inverseFunction <- family(mod1)$linkinv

# transform predicted data to the natural scale
newData <- newData %>%
  mutate(
    nSpecies = inverseFunction(nSpecies_LP),
    ymin = inverseFunction(lowerCI_LP),
    ymax = inverseFunction(upperCI_LP)
  )
```

Now we can add these to the plot:

```
ggplot(roundabouts, aes(
  x = area, y = nSpecies,
  colour = habitatType
)) +
  geom_ribbon(data = newData, aes(
    x = area, ymin = ymin, ymax = ymax,
    fill = habitatType, alpha = 0.3
  )) +
  geom_smooth(data = newData, stat = "identity") +
  geom_point() +
  guides(alpha = FALSE) + # Remove the alpha legend
  NULL
```


nSpecies
20
15
10
5
0
0
10
20
30
40
50
area
habitatType
complex
simple

Part V

Solutions

Chapter 23

Exercise Solutions

These are the solutions to the exercises used in the course.

23.1 Californian bird diversity

1. First import the data. Check that the columns look as they should. (e.g. use `summary` or `str` functions). Tip: use the "Wizard" in RStudio to guide you.

```
df <- read.csv("CourseData/suburbanBirds.csv")
```

2. What is the mean, minimum, and maximum number of species seen? (there is more than one way to do this)*

```
mean(df$nSpecies)
```

```
## [1] 9.647059
```

```
min(df$nSpecies)
```

```
## [1] 3
```

```
max(df$nSpecies)
```

```
## [1] 15
```

```
range(df$nSpecies)
```

```
## [1]  3 15
```

```
summary(df$nSpecies)
```

```
##    Min. 1st Qu.  Median    Mean 3rd Qu.    Max.
##   3.000   6.000  11.000   9.647  13.000  15.000
```

3. How old are the youngest and oldest suburbs? (hint: the survey was carried out in 1975, do the math!)

```
1975 - min(df$Year)
```

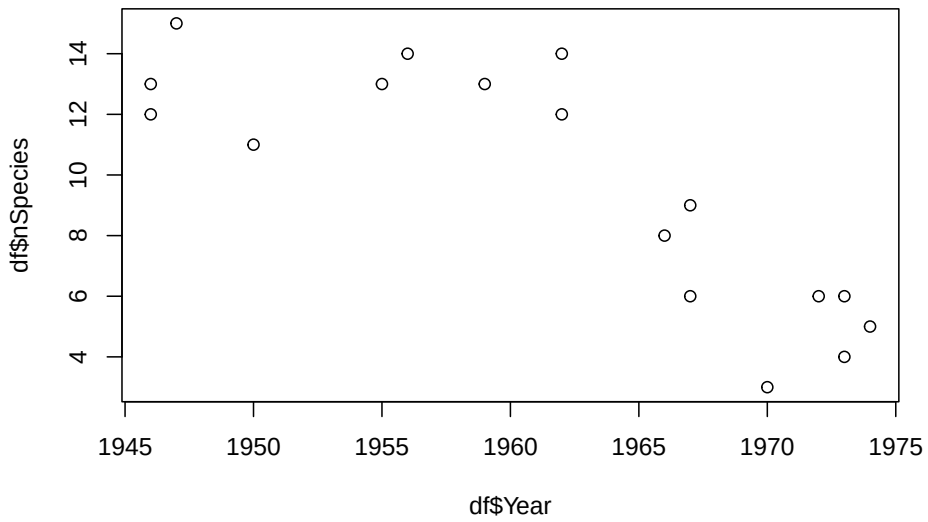
```
## [1] 29
```

```
1975 - max(df$Year)
```

```
## [1] 1
```

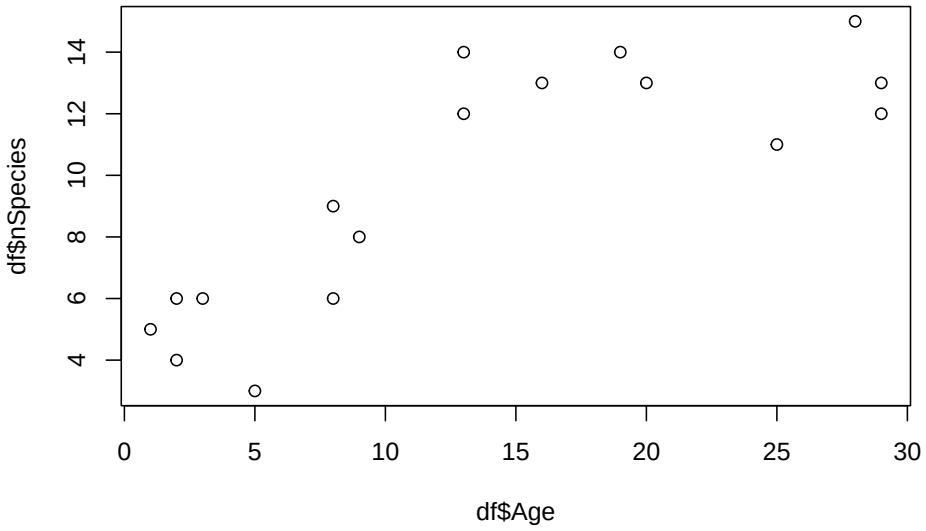
4. Plot the relationship between `Year` and `nSpecies` as a scatterplot using base-R graphics (using the `plot` function).

```
plot(df$Year, df$nSpecies)
```



5. The pattern might be easier to see if you could replace `YearBuilt` with suburb age. Create a new vector in your data frame for this variable (e.g. `df$Age <- 1975 - Year`). Replot your results.

```
df$Age <- 1975 - df$Year
plot(df$Age, df$nSpecies)
```



6. What do the data show? What might be the mechanisms for the patterns you see? Do they match your expectations?

If you recall that the average species richness pre-development was about 3.5 species, the data show that suburban development is actually good for bird species. This could be surprising, but a possible explanation is that the gardens, parks, trees etc. that come with development represent additional habitats that would not normally be there. Therefore, these areas attract new species.

7. Export your plots and paste them into a Word Document.

You can do this with several methods. My favourite *quick* method is to click the Export button > Copy to Clipboard, resize the plot so it looks nice, then click Copy Plot. Finally, paste into Word with Ctrl (or Cmd) + V.

23.2 Wrangling the Amniote Life History Database

1. When you have imported the data, use `dim` to check the dimensions of the whole data frame (you should see that there are 36 columns and 21322 rows). Use `names` to look at the names of all columns in the data in `amniote`.

```
amniote <- read.csv("CourseData/Amniote_Database_Aug_2015.csv",
  na.strings = "-999"
)
```

```
dim(amniote)
```

```
## [1] 21322    36
```

```
names(amniote)
```

```
##  [1] "class"
##  [2] "order"
##  [3] "family"
##  [4] "genus"
##  [5] "species"
##  [6] "subspecies"
##  [7] "common_name"
##  [8] "female_maturity_d"
##  [9] "litter_or_clutch_size_n"
## [10] "litters_or_clutches_per_y"
## [11] "adult_body_mass_g"
## [12] "maximum_longevity_y"
## [13] "gestation_d"
## [14] "weaning_d"
## [15] "birth_or_hatching_weight_g"
## [16] "weaning_weight_g"
## [17] "egg_mass_g"
## [18] "incubation_d"
## [19] "fledging_age_d"
## [20] "longevity_y"
## [21] "male_maturity_d"
## [22] "inter_litter_or_interbirth_interval_y"
## [23] "female_body_mass_g"
## [24] "male_body_mass_g"
## [25] "no_sex_body_mass_g"
## [26] "egg_width_mm"
## [27] "egg_length_mm"
## [28] "fledging_mass_g"
## [29] "adult_svl_cm"
## [30] "male_svl_cm"
## [31] "female_svl_cm"
## [32] "birth_or_hatching_svl_cm"
## [33] "female_svl_at_maturity_cm"
## [34] "female_body_mass_at_maturity_g"
## [35] "no_sex_svl_cm"
## [36] "no_sex_maturity_d"
```

2. We are interested in longevity (lifespan) and body size and reproductive effort and how this might vary depending on the taxonomy (specifically, with Class). Use `select` to pick relevent columns of the dataset and discard the others. Call the new data frame `x`. The relevant columns are the taxonomic variables (`class`, `species`) and `longevity_y`, `litter_or_clutch_size_n`, `litters_or_clutches_per_y`, and `adult_body_mass_g`.

```
x <- amniote %>%
  select(
    class, genus, species,
    longevity_y, adult_body_mass_g,
    litter_or_clutch_size_n, litters_or_clutches_per_y
  )
```

3. Take a look at the first few entries in the `species` column. You will see that it is only the *epithet*, the second part of the *Genus_species* name, that is given.
   Use `mutate` and `paste` to convert the `species` column to a *Genus_species* by pasting the data in `genus` and `species` together. To see how this works, try out the following command, `paste(1:3, 4:6)`. After you have created the new column, remove the `genus` column (using `select` and `-genus`).

```
x <- x %>%
  mutate(species = paste(genus, species)) %>%
  select(-genus)
head(x)
```

```
##   class               species longevity_y adult_body_mass_g
## 1  Aves Accipiter albogularis          NA           251.500
## 2  Aves      Accipiter badius          NA           140.000
## 3  Aves     Accipiter bicolor          NA           345.000
## 4  Aves  Accipiter brachyurus          NA           142.000
## 5  Aves    Accipiter brevipes          NA           203.500
## 6  Aves Accipiter castanilius          NA           159.375
##   litter_or_clutch_size_n litters_or_clutches_per_y
## 1                      NA                        NA
## 2                    3.25                         1
## 3                    2.70                        NA
## 4                      NA                        NA
## 5                    4.00                         1
## 6                      NA                        NA
```

4. What is the longest living species in the record? Use `arrange` to sort the data from longest to shortest longevity (`longevity_y`), and then look at the top of the file using `head` to find out. (hint: you will need to use reverse sort (`-`)). Cut and paste the species name into Google to find out more!

```
x <- x %>% arrange(-longevity_y)
head(x)
```

```
##      class                 species longevity_y adult_body_mass_g
## 1 Reptilia Chelonoidis duncanensis       177.0               NA
## 2 Reptilia  Aldabrachelys gigantea       152.0           117200
## 3 Reptilia          Testudo graeca       127.0             1430
## 4 Mammalia            Homo sapiens       122.5            62035
## 5 Mammalia   Balaenoptera physalus        95.0         38800000
## 6 Mammalia            Orcinus orca        90.0          4300000
##   litter_or_clutch_size_n litters_or_clutches_per_y
## 1                      NA                       NA
## 2                    13.5                 2.000000
## 3                     5.0                 3.200993
## 4                     1.0                 0.485000
## 5                     1.0                 0.400000
## 6                     1.0                 0.210000
```

5. Do the same thing but this time find the shortest lived species.

```
x <- x %>% arrange(longevity_y)
head(x)
```

```
##      class              species longevity_y adult_body_mass_g
## 1 Mammalia    Lepus nigricollis  0.08333333          2196.875
## 2 Mammalia  Notoryctes caurinus  0.08333333            34.000
## 3 Mammalia Allactaga balikunica  0.08333333                NA
## 4 Mammalia    Allactaga bullata  0.08333333                NA
## 5 Mammalia       Geomys pinetis  0.08333333           195.750
## 6 Mammalia          Mus sorella  0.08333333            12.535
##   litter_or_clutch_size_n litters_or_clutches_per_y
## 1                    1.59                    7.150
## 2                    1.50                       NA
## 3                    2.52                       NA
## 4                    2.52                       NA
## 5                    1.77                    1.865
## 6                    5.20                    3.000
```

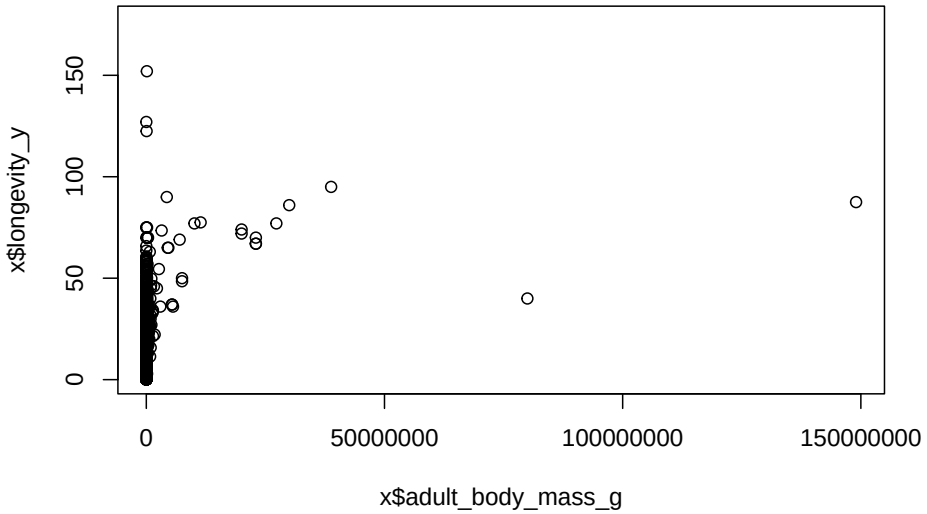
6. Use `summarise` and `group_by` to make a table summarising `min`, `median` and `max` life spans (`longevity_y`) for the three taxonomic classes in the database. Remember that you need to tell R to remove the `NA` values using a `na.rm = TRUE` argument.

```
x %>%
  group_by(class) %>%
  summarise(
    min = min(longevity_y, na.rm = TRUE),
    median = median(longevity_y, na.rm = TRUE),
    max = max(longevity_y, na.rm = TRUE)
  )
```

```
## # A tibble: 3 x 4
##   class       min median   max
##   <chr>     <dbl>  <dbl> <dbl>
## 1 Aves     0.75    12.5    75
## 2 Mammalia 0.0833   8.67  122.
## 3 Reptilia 0.2     11.0   177
```

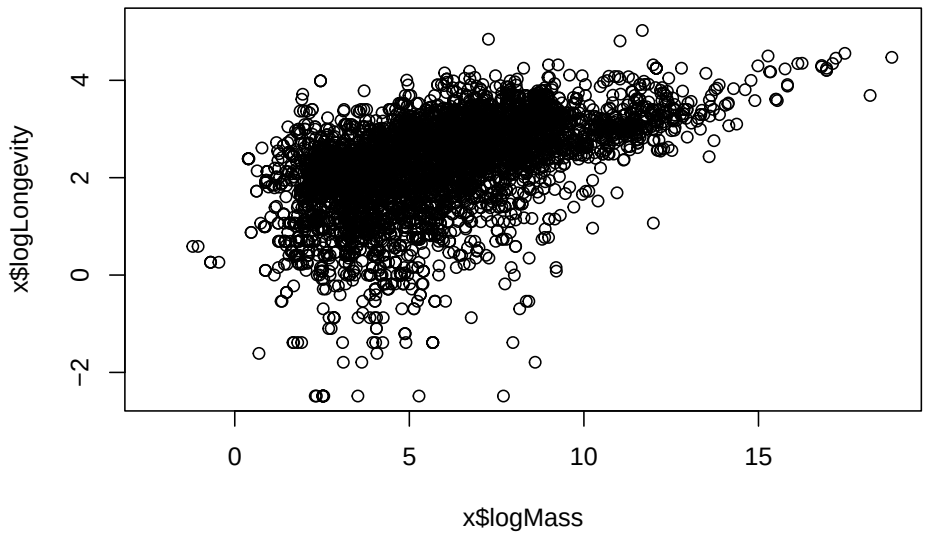
7. Body size is thought to be associated with life span. Let's treat that as a hypothesis and test it graphically. Sketch what would the graph would look like if the hypothesis were true, and if it was false. Plot `adult_body_mass_g` vs. `longevity_y` (using base R graphics). You should notice that this looks a bit messy.

```
plot(x$adult_body_mass_g, x$longevity_y)
```



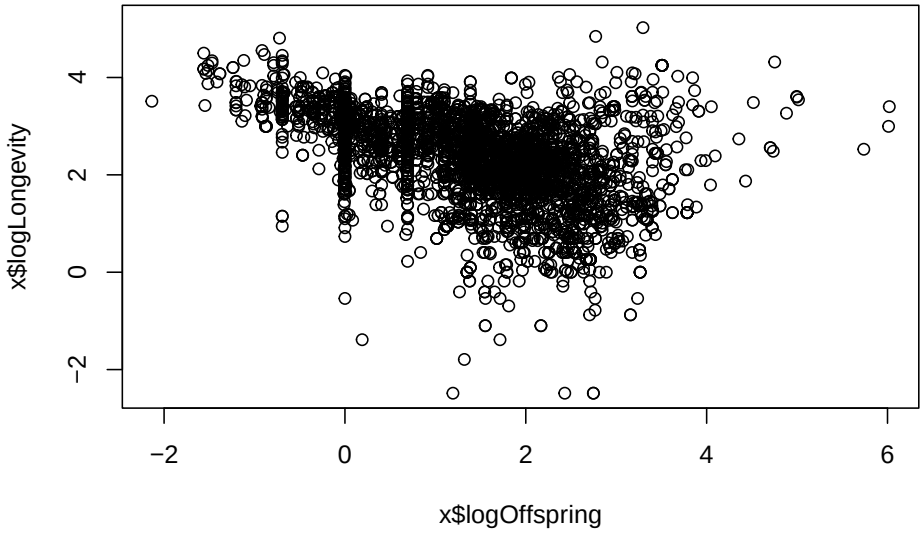
8. Use `mutate` to create a new `log`-transformed variables, `logMass` and `logLongevity`. Use these to make a "log-log" plot. You should see that makes the relationship more linear, and easier to "read".

```
x <- x %>%
  mutate(
    logMass = log(adult_body_mass_g),
    logLongevity = log(longevity_y)
  )

plot(x$logMass, x$logLongevity)
```



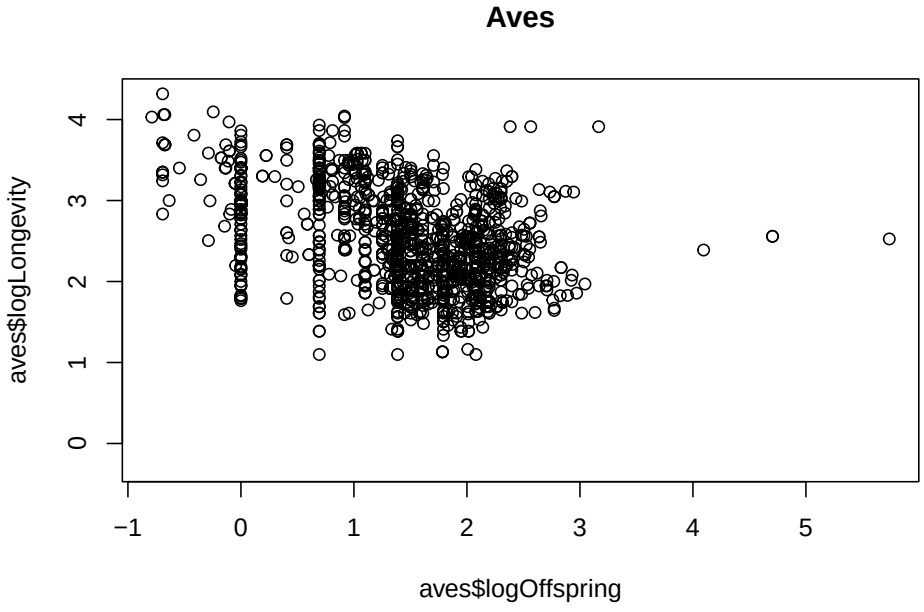
9. Is there a trade-off between reproductive effort and life span? Think about this as a hypothesis - sketch what would the graph would look like if that were true, and if it was false. Now use the data to test that hypothesis: Use `mutate` to create a variable called `logOffspring` which is the logarithm of number of litters/clutches per year multiplied by the number of babies in each litter/clutch . Then plot `logOffspring` vs. `logLongevity`.

```
x <- x %>%
  mutate(logOffspring = log(
    litter_or_clutch_size_n * litters_or_clutches_per_y
  ))

plot(x$logOffspring, x$logLongevity)
```

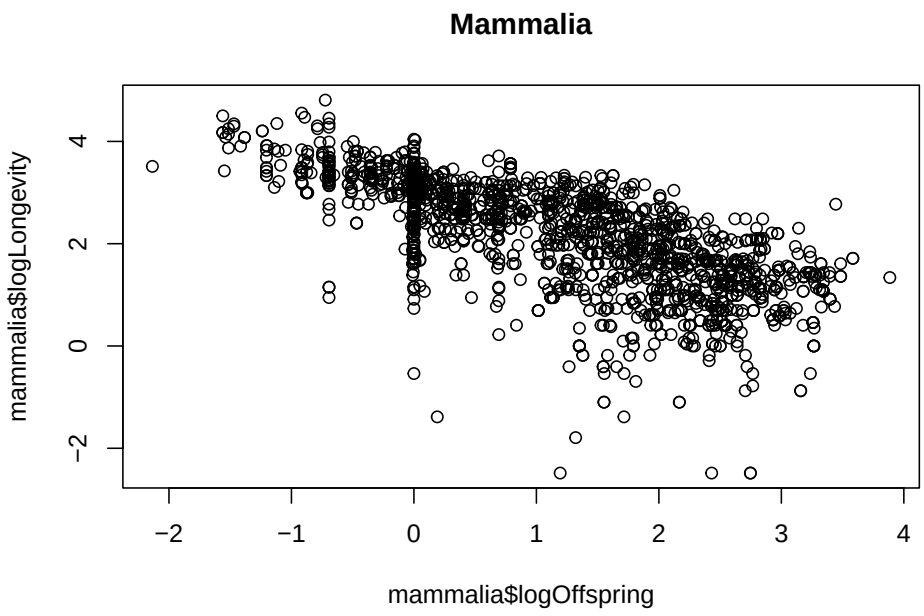


10. To answer the final question (differences between taxonomic classes) you could now use `filter` to subset to particular classes and repeat the plot to see whether the relationships holds universally.

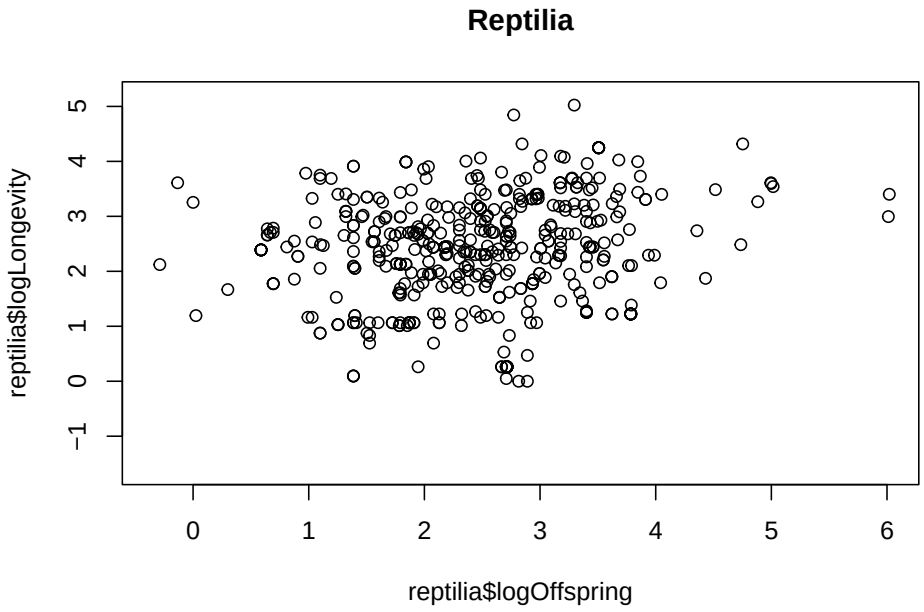
```
aves <- x %>%
  filter(class == "Aves")

plot(aves$logOffspring, aves$logLongevity)
title("Aves")
```



```
mammalia <- x %>%
  filter(class == "Mammalia")

plot(mammalia$logOffspring, mammalia$logLongevity)
title("Mammalia")
```




```
reptilia <- x %>%
  filter(class == "Reptilia")

plot(reptilia$logOffspring, reptilia$logLongevity)
title("Reptilia")
```



23.3 Temperature effects on egg laying dates

1. Import the data and take a look at it with `head` or `str`.

```
eggDates <- read.csv("CourseData/eggDates.csv")
head(eggDates)
```

```
##   boxNumber y2013 y2014 y2016 y2017 y2018 y2019
## 1         1   116   103    NA   107   111    NA
## 2         2    NA    NA    NA   114   118    NA
## 3         3    NA   102   108    NA    NA    NA
## 4         4   121   103   121   155   111   110
## 5         5   135   100   108   102   106   108
## 6         6   122   113   122    NA   124   149
```

2. Use `pivot_longer` to reformat the data. This might take a bit of trial and error - don't give up!

Maybe this will help: The first argument in the `pivot_longer` command (`cols`) tells R which columns contain the data you are interested in (in this case, these are `y2013`,`y2014` etc). Then the `names_to` argument tells R what you want to name the new column from this data (in this case, `Year`). Then, the `values_to` argument tells R what the data column should be called (e.g. `Day`). In addition, there is a useful argument called `names_prefix` that will remove the part of the column name (e.g. the `y` of `y2013`)

You should also make sure that the `Year` column is recognised as being a numeric variable rather than a character string. You can do this by adding a command using `mutate` and `as.numeric`, like this `mutate(Year = as.numeric(Year))`

You should end up with a dataset with three columns as described above.

```
eggDates <- eggDates %>% pivot_longer(
  cols = starts_with("y"),
  names_to = "Year", values_to = "day"
)

head(eggDates)
```

```
## # A tibble: 6 x 3
##   boxNumber Year    day
##       <int> <chr> <int>
## 1         1 y2013   116
## 2         1 y2014   103
## 3         1 y2016    NA
## 4         1 y2017   107
## 5         1 y2018   111
## 6         1 y2019    NA
```

3. Ensure that year is coded as numeric variable using `mutate`. [Hint, you can use the command `as.numeric`, but first remove the "y" in the name using `gsub`].

```
eggDates <- eggDates %>%
  mutate(Year = gsub(pattern = "y", replacement = "", Year)) %>%
  mutate(Year = as.numeric(Year))
head(eggDates)
```

```
## # A tibble: 6 x 3
##   boxNumber  Year   day
##       <int> <dbl> <int>
## 1         1  2013   116
## 2         1  2014   103
## 3         1  2016    NA
## 4         1  2017   107
## 5         1  2018   111
## 6         1  2019    NA
```

4. Calculate the mean egg date per year using `summarise` (remember to `group_by` the year first). Take a look at the data.

```
meanEgg <- eggDates %>%
  group_by(Year) %>%
  summarise(meanEggDate = mean(day, na.rm = TRUE))

meanEgg
```

```
## # A tibble: 6 x 2
##    Year meanEggDate
##   <dbl>       <dbl>
## 1  2013        125.
## 2  2014        108.
## 3  2016        115.
## 4  2017        112.
## 5  2018        117.
## 6  2019        111.
```

Preparing the weather data

5. Import the weather data and take a look at it with `head` or `str`.

```
weather <- read.csv("CourseData/AarslevTemperature.csv")
str(weather)
```

```
## 'data.frame':    3291 obs. of  4 variables:
##  $ YEAR : int  2012 2012 2012 2012 2012 2012 2012 2012 2012 2012 ...
##  $ MONTH: int  4 4 4 4 4 4 4 4 4 4 ...
##  $ DAY  : int  11 12 13 14 15 16 17 18 19 20 ...
##  $ TEMP : num  8.1 6.6 5.9 6.2 6.5 4.3 4.3 7 8.9 6.5 ...
```

6. Use `filter` subset to the months of interest (February-April) and then `summarise` the data to calculate the mean temperature in this period (remember to `group_by` year). Look at the data. You should end up with a dataset with two columns - `YEAR` and `meanSpringTemp`.

```
weather <- weather %>%
  filter(MONTH %in% 2:4) %>%
  group_by(YEAR) %>%
  summarise(meanAprilTemp = mean(TEMP))

head(weather)
```

```
## # A tibble: 6 x 2
##    YEAR meanAprilTemp
##   <int>         <dbl>
```

```
## 1  2012          7.86
## 2  2013          1.60
## 3  2014          6.03
## 4  2015          4.63
## 5  2016          4.50
## 6  2017          4.77
```

Bringing it together

7. Join the two datasets together using `left_join`. You should now have a dataset with columns `nestNumber`, `Year`, `dayNumber` and `meanAprilTemp`

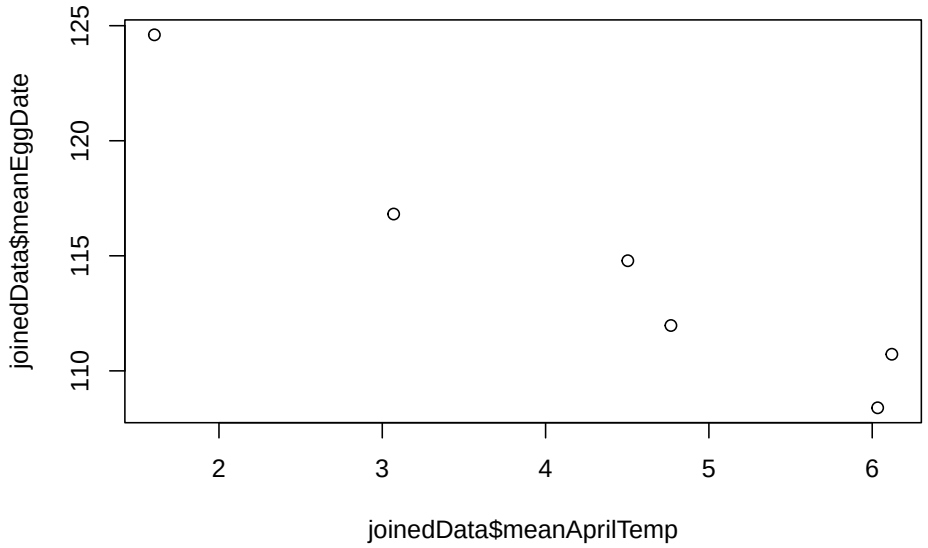
```
joinedData <- left_join(meanEgg, weather, c("Year" = "YEAR"))
head(joinedData)
```

```
## # A tibble: 6 x 3
##    Year meanEggDate meanAprilTemp
##   <dbl>       <dbl>         <dbl>
## 1  2013        125.          1.60
## 2  2014        108.          6.03
## 3  2016        115.          4.50
## 4  2017        112.          4.77
## 5  2018        117.          3.07
## 6  2019        111.          6.12
```

Plot the data

8. plot a graph of `meanAprilTemp` on the x-axis and `dayNumber` on the y-axis.

```
plot(joinedData$meanAprilTemp, joinedData$meanEggDate)
```



Now you should be able to answer the question we started with: is laying date associated with spring temperatures? Yes, there looks to be a negative relationship between temperature and egg laying date.

23.4 Virtual dice

Let's try the same kind of thing with the roll of (virtual) dice.

Here's how to do one roll of the dice:

```
diceRoll <- 1:6
sample(diceRoll, 1)
```

[1] 3

1) Simulate 10 rolls of the dice, and make a table of results.

```
result <- sample(diceRoll, 10, replace = TRUE)
table(result)
```

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

Your table will probably look different to this, because it is a random process. You may notice that some numbers in the table are missing if some numbers were never rolled by our virtual dice.

2) Now simulate 90 rolls of the dice, and plot the results as a bar plot using `geom_bar` in `ggplot`. Add a horizontal line using `geom_abline` to show the **expected** result based on what you know about probability.

```
n <- 90
result <- data.frame(result = sample(diceRoll, n, replace = TRUE))

ggplot(result, aes(x = result)) +
  geom_bar() +
  geom_abline(intercept = n / 6, slope = 0)
```

3) Try adjusting the code to simulate dice rolls with small (say, 30) and large (say, 600, or 6000, or 9000) samples. Observe what happens to the proportions, and compare them to the expected value. What does this tell you about the importance of sample size when trying to estimate real phenomena?

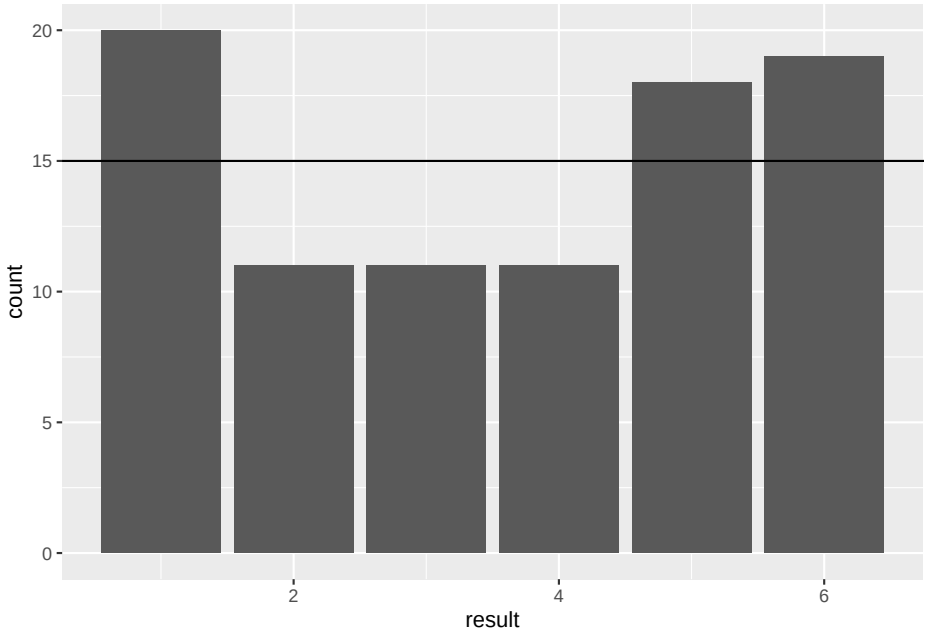


Figure 23.1: Barplot of 90 simulated dice throws

You only need to edit the `n <- 90` line in the code above

The main message here is that as sample size increases you are more likely to obtain a good estimate of the true value of the phenomenon. You may also notice that, what would be considered a good sample size for the coin flipping (i.e. it recovers the true probability of 0.5 reasonably well) is NOT adequate for getting a good estimate of the probabilities for the dice.

This is because of the different number of possibilities: as the range of possible outcomes increases, the sample size requirements increase. In other words, choosing a good sample size is context-dependent.

23.5 Sexual selection in Hercules beetles

1. What is your null hypothesis?

Null Hypothesis - There is no difference in the widths of the species.

2. What is your alternative hypothesis?

Alternative Hypothesis - Males have larger widths than females.

3. Import the data.

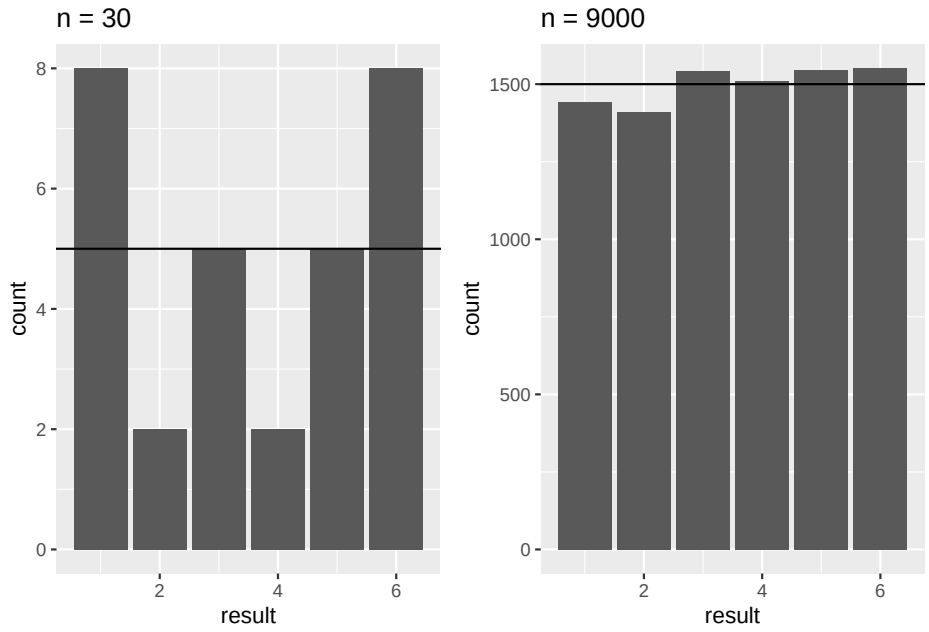


Figure 23.2: Barplots of 30 and 9000 simulated dice throws

```
hercules <- read.csv("CourseData/herculesBeetle.csv")
```

4. Calculate the mean for each sex (either using `tapply` or using `dplyr` tools)

```
# With dplyr
hercules %>%
  group_by(sex) %>%
  summarise(mean = mean(width))
```

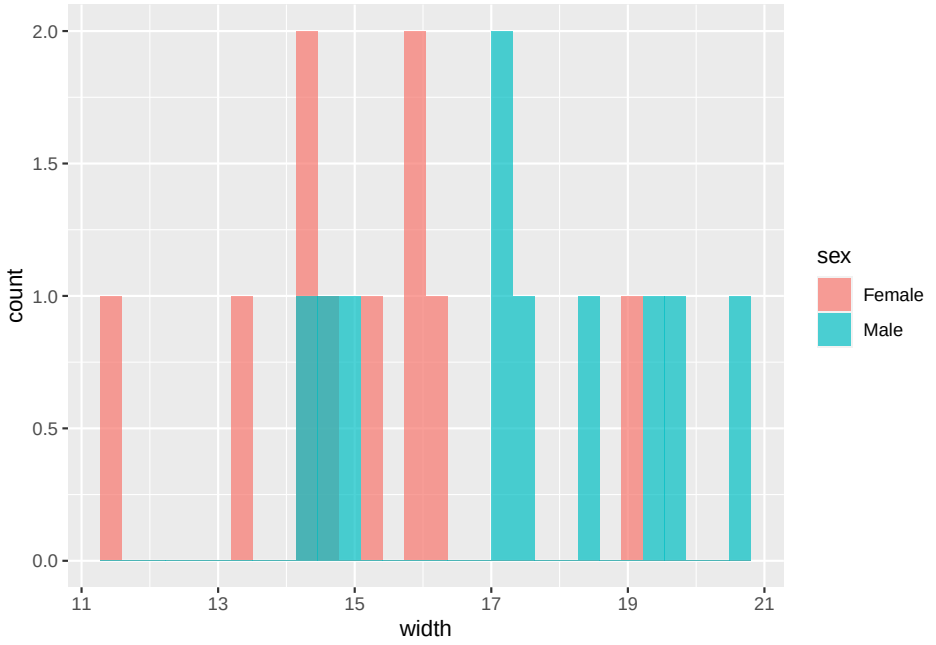
```
## # A tibble: 2 x 2
##   sex     mean
##   <chr>  <dbl>
## 1 Female  15.0
## 2 Male    17.4
```

```
# with tapply
tapply(hercules$width, hercules$sex, mean)
```

```
##   Female     Male
## 15.02825 17.36568
```

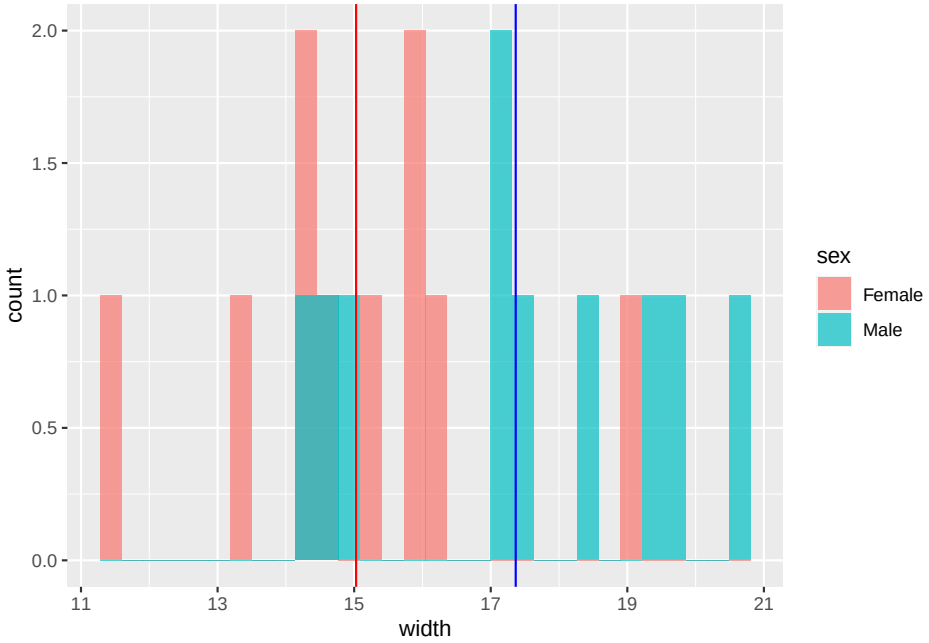
5. Plot the data as a histogram.

```r
# Let's look at the male and female data
(plot1 <- ggplot(hercules, aes(x = width, fill = sex)) +
  geom_histogram(position = "identity", alpha = .7))
```



6. Add vertical lines to the plot to indicate the mean values.

```r
hercules %>%
  group_by(sex) %>%
  summarise(mean = mean(width)) %>%
  pull(mean) -> meanVals

plot1 + geom_vline(xintercept = meanVals, colour = c("red", "blue"))
```



7. Now calculate the difference between the mean values using `dplyr` tools, or `tapply`.

```
# with dplyr
hercules %>%
  group_by(sex) %>%
  summarise(mean = mean(width)) %>%
  pull(mean) %>%
  diff() -> observedDiff
observedDiff
```

```
## [1] 2.337433
```

```
# with tapply
diff(as.vector(tapply(hercules$width, hercules$sex, mean)))
```

```
## [1] 2.337433
```

8. Use `sample` to randomise the sex column of the data, and recalculate the difference between the mean.

```
# with dplyr
hercules %>%
  mutate(sex = sample(sex)) %>%
```

```
  group_by(sex) %>%
  summarise(mean = mean(width)) %>%
  pull(mean) %>%
  diff()
```

```
## [1] -1.900753
```

```
# with tapply
diff(as.vector(tapply(hercules$width, sample(hercules$sex), mean)))
```

```
## [1] 0.3205587
```

9. Use `replicate` to repeat this 10 times (to ensure that you code works).

```
# with dplyr
replicate(
  10,
  hercules %>%
    mutate(sex = sample(sex)) %>%
    group_by(sex) %>%
    summarise(mean = mean(width)) %>%
    pull(mean) %>%
    diff()
)
```

```
##  [1]  0.3885126 -0.5711667 -0.2898130 -0.3766535 -0.4209082 -0.4678595
##  [7] -2.1980346 -1.5961324  0.8836404  1.2863489
```

```
# with tapply
replicate(
  10,
  diff(as.vector(tapply(
    hercules$width,
    sample(hercules$sex), mean
  )))
)
```

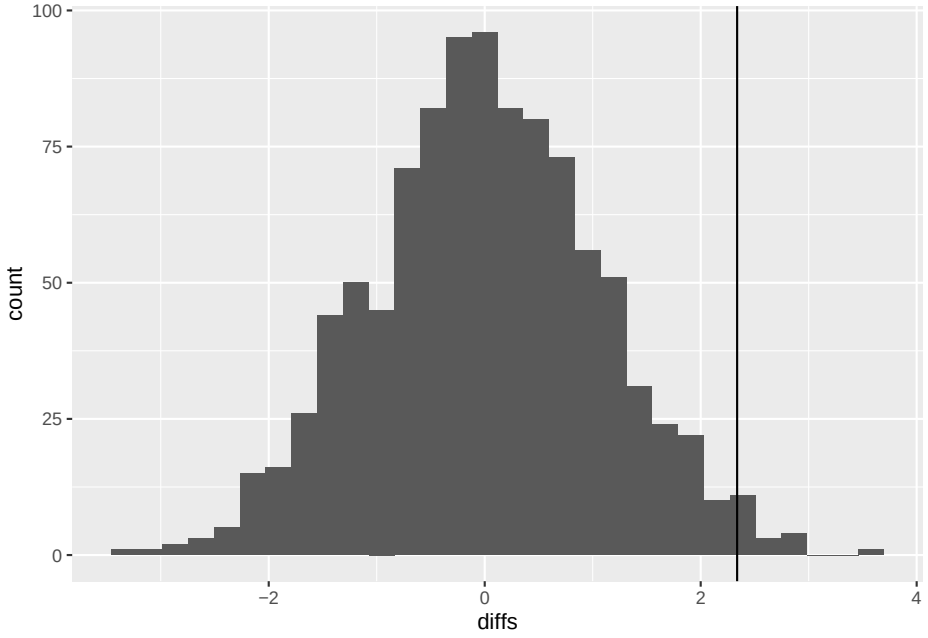
```
##  [1]  1.2267611 -0.2954281  0.3783011 -0.6666253 -1.4024738  0.6233728
##  [7] -0.5334274  0.2076405 -0.4760695 -0.6967478
```

10. When your code is working, use `replicate` again, but this time with 1000 replicates and pass the results into a data frame.

```r
# with dplyr
diffs <- data.frame(
  diffs =
    replicate(
      1000,
      hercules %>%
        mutate(sex = sample(sex)) %>%
        group_by(sex) %>%
        summarise(mean = mean(width)) %>%
        pull(mean) %>%
        diff()
    )
)

# with tapply
diffs <- data.frame(
  diffs =
    replicate(
      1000,
      diff(as.vector(
        tapply(hercules$width, sample(hercules$sex), mean)
      ))
    )
)
```

11. Use `ggplot` to plot the null distribution you have just created, and add the observed difference.

```r
ggplot(diffs, aes(x = diffs)) +
  geom_histogram() +
  geom_vline(xintercept = observedDiff)
```



12. Obtain the p-value for the hypothesis test described above. (1) how many of the shuffled differences are more extreme than the observed distribution (2) what is this expressed as a proportion of the number of replicates.

```
sum(diffs$diffs >= observedDiff)
```

```
## [1] 14
```

```
sum(diffs$diffs >= observedDiff) / 1000
```

```
## [1] 0.014
```

13. Summarise your result as in a report. Describe the method, followed by the result and conclusion.

"I used a randomisation test to estimate the significance of the observed difference of 2.337 (mean values: female=17.366; male = 15.028) in mean widths of the sexes. To do this I generated a null distribution of differences between sexes using 1000 replicates. I found that only 14 of the differences in the null distribution were as extreme as the observed difference. Thus the p-value is 0.014: I therefore reject the null hypothesis that there is no difference between the sexes and accept the alternative hypothesis that males are significantly larger than females."

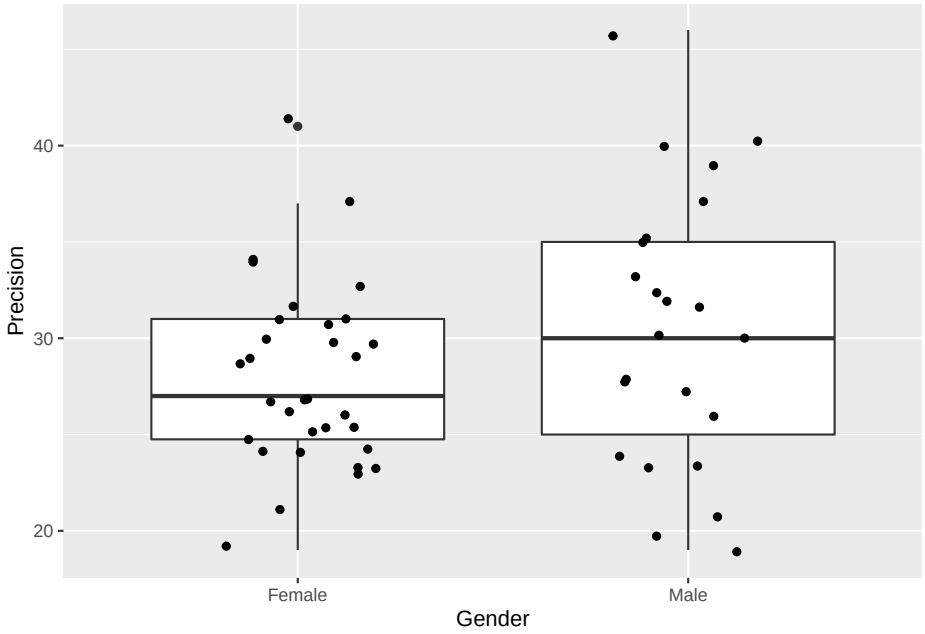
23.6 Sex differences in fine motor skills

Some people have suggested that there might be sex differences in fine motor skills in humans. Use the data collected on the class to address this topic using t-tests. The relevant data set is called `classData.csv`, and the columns of interest are `Gender` and `Precision`.

Carry out a two-sample t-test.

1) Plot the data (e.g. with a box plot, or histogram)

```
classData <- read.csv("CourseData/classData.csv")

ggplot(classData, aes(x = Gender, y = Precision)) +
  geom_boxplot() +
  geom_jitter(width = 0.2)
```



2) Formulate null and alternative hypotheses.

Null hypotheses - the differences in precision between male and female are due to chance alone. Alternative hypothesis - there is a significant difference between male and female precision scores.

3) Use the `t.test` function to do the test.

```
t.test(Precision ~ Gender, data = classData)
```

```
##
##  Welch Two Sample t-test
##
## data:  Precision by Gender
## t = -1.4413, df = 35.759, p-value = 0.1582
## alternative hypothesis: true difference in means between group Female and group Ma
## 95 percent confidence interval:
##  -5.936817  1.004752
## sample estimates:
## mean in group Female   mean in group Male
##             27.96875             30.43478
```

4) Write a sentence or two describing the results.

There was no significant difference in mean precision between the two genders (t-test: t = -1.44, df = 35.76, p = 0.158). The 95% confidence interval for the difference between genders overlapped 0 (95% CI = -5.937-1.005). I therefore fail to reject the null hypothesis that the observed differences are due to chance alone.

23.7 Therapy for anorexia

A study was carried out looking at the effect of cognitive behavioural therapy on weight of people with anorexia. Weight was measured in week 1 and again in week 8. Use a paired t-test to assess whether the treatment is effective.

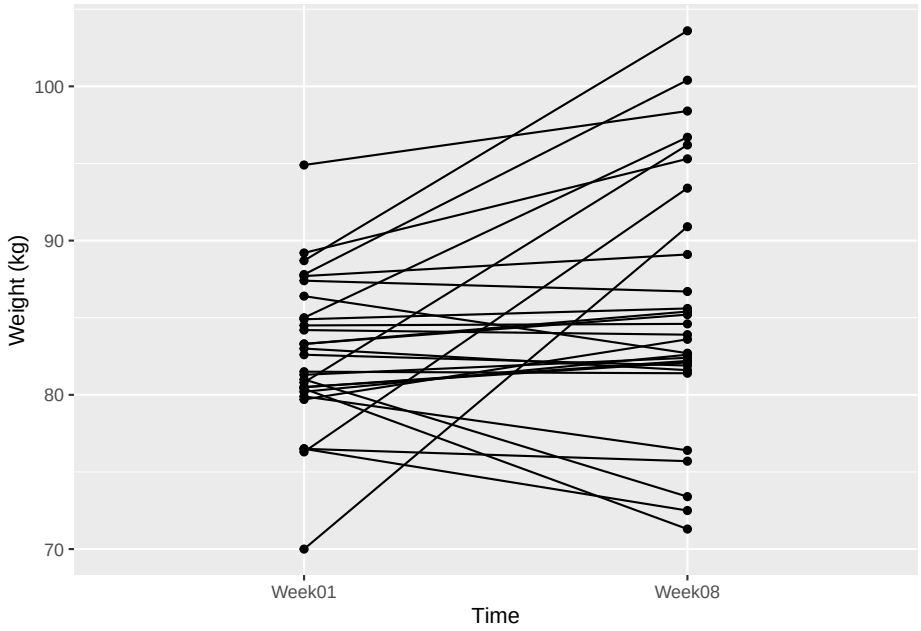
The data is called `anorexiaCBT.csv`

The data are in "wide format". You may wish to convert it to "long format" depending on how you use the data. You can do that with the `pivot_longer` function, which rearranges the data:

1) Plot the data (e.g. with an interaction plot like Figure 14.5)

```
anorexiaCBT <- read.csv("CourseData/anorexiaCBT.csv",
  header = TRUE
)

anorexiaCBT_long <- anorexiaCBT %>%
  pivot_longer(
    cols = starts_with("Week"), names_to = "time",
    values_to = "weight"
  )
```

```
ggplot(anorexiaCBT_long, aes(
  x = time, y = weight,
  group = Subject
)) +
  geom_line() +
  geom_point() +
  xlab("Time") +
  ylab("Weight (kg)")
```



2) Formulate a null and alternative hypothesis.

Null = The difference in weight between the two times is no different than random chance. Alternative Hypothesis = There is a significant change in weight between the two time points.

3) Use `t.test` to conduct a *paired* t-test.

The method here depends on whether you use the "long" data or not:

```
t.test(anorexiaCBT$Week01, anorexiaCBT$Week08, paired = TRUE)
```

```
##
##  Paired t-test
##
## data:  anorexiaCBT$Week01 and anorexiaCBT$Week08
```

```
## t = -2.2156, df = 28, p-value = 0.03502
## alternative hypothesis: true difference in means is not equal to 0
## 95 percent confidence interval:
##  -5.7869029 -0.2268902
## sample estimates:
## mean of the differences
##               -3.006897
```

or

```
anorexiaCBT_long <- anorexiaCBT_long %>%
  arrange(Subject, time)

t.test(weight ~ time, data = anorexiaCBT_long, paired = TRUE)
```

```
##
##  Paired t-test
##
## data:  weight by time
## t = -2.2156, df = 28, p-value = 0.03502
## alternative hypothesis: true difference in means is not equal to 0
## 95 percent confidence interval:
##  -5.7869029 -0.2268902
## sample estimates:
## mean of the differences
##               -3.006897
```

4) Write a couple of sentences to report your result.

```
x <- t.test(anorexiaCBT$Week01, anorexiaCBT$Week08, paired = TRUE)
```

There was a significant difference in weight of -3.007kg between week 1 and week 8 (t.test: t = -2.22, df = 28.00, p = 0.035). The 95% confidence interval for the difference between weeks was between -5.787 and -0.227. Therefore, I reject the null hypotheses, that the difference is due to chance alone, and accept the alternative hypothesis.

23.8 Compare t-tests with randomisation tests

Try re-fitting some of these tests as randomisation tests (or analyse the randomisation test data using `t.test`). Do they give approximately the same results?
Then try answering the question - "*are people who prefer dogs taller than those who prefer cats?*" using the `classData.csv`. Can you think of any problems with this analysis?

The problem with the analysis is that it is "confounded". That is to say, gender is correlated with height, so you would not be sure whether any difference you found would be due to height, or gender.

23.9 Apple tree crop yield

Import the data (`apples.csv`) and analyse it using the techniques you have learned in the ANOVA lecture, and the previous worksheet, to answer the question "What is the effect of tree spacing on apple yields?"

1. Import and look at the data (e.g. `summary` or `str` or `head`)

```
apples <- read.csv("CourseData/apples.csv")
summary(apples)
```

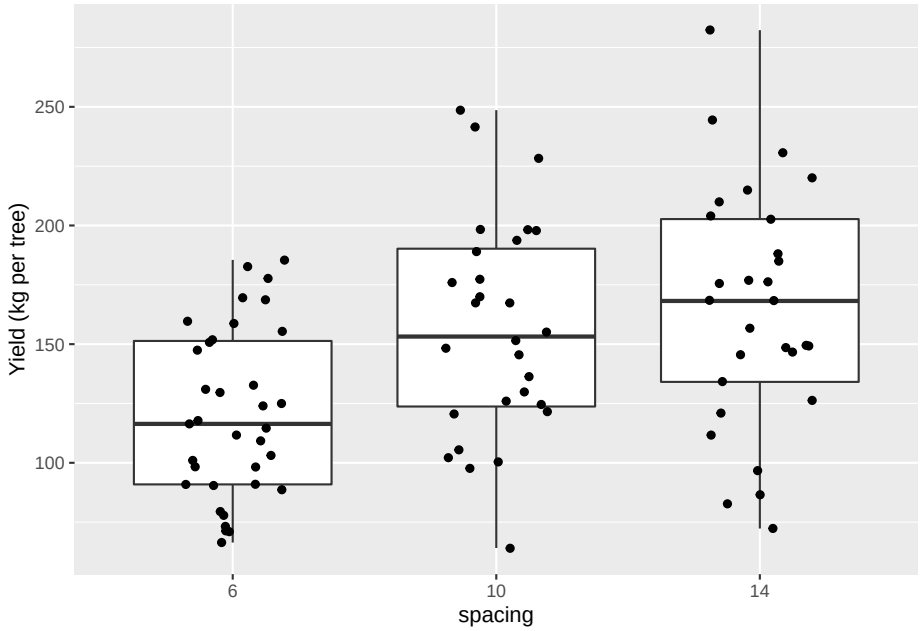
```
##     spacing       yield
##  Min.   : 6   Min.   : 64.1
##  1st Qu.: 6   1st Qu.:108.2
##  Median :10   Median :147.1
##  Mean   :10   Mean   :145.4
##  3rd Qu.:14   3rd Qu.:176.5
##  Max.   :14   Max.   :282.3
##               NA's   :28
```

2. Ensure that the explanatory variable (`spacing`) is defined as a categorical variable (i.e. a "factor", in R-speak). You can use `mutate` and `as.factor` functions for this.

```
apples <- apples %>%
  mutate(spacing = as.factor(spacing))
```

3. Plot the data using `ggplot` (a box plot with (optionally) added jittered points would be good).

```
ggplot(apples, aes(x = spacing, y = yield)) +
  geom_boxplot() +
  geom_jitter(width = 0.2) +
  ylab("Yield (kg per tree)")
```

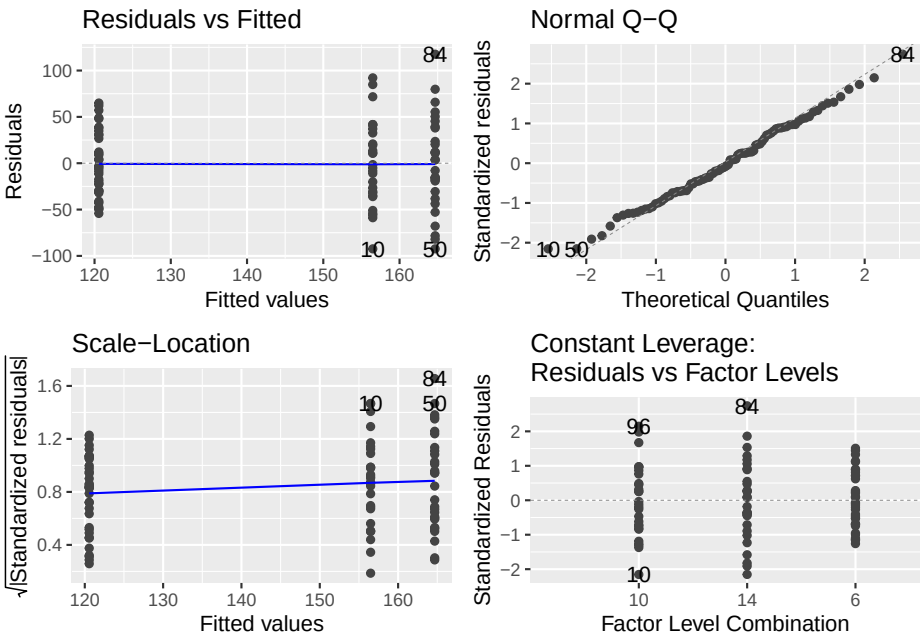


4. Fit an ANOVA model using `lm`.

```
apple_mod <- lm(yield ~ spacing, data = apples)
```

5. Check the model using a diagnostic plot (i.e. using `autoplot` from the `ggfortify` package).

```
library(ggfortify)
autoplot(apple_mod)
```



6. Use `anova` to get the ANOVA summary.

```
anova(apple_mod)
```

```
## Analysis of Variance Table
##
## Response: yield
##           Df Sum Sq Mean Sq F value    Pr(>F)
## spacing    2  35801 17900.3  9.3851 0.0002003 ***
## Residuals 89 169750  1907.3
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

7. You should see that there are differences among treatments. But where are those differences? Use `summary` on your model to find out.

```
summary(apple_mod)
```

```
##
## Call:
## lm(formula = yield ~ spacing, data = apples)
##
## Residuals:
##     Min      1Q  Median      3Q     Max
## -92.389 -30.577  -3.516  33.192 117.628
```

```
##
## Coefficients:
##             Estimate Std. Error t value             Pr(>|t|)
## (Intercept)  120.566      7.382  16.332 < 0.0000000000000002 ***
## spacing10     35.924     11.073   3.244             0.001659 **
## spacing14     44.107     10.966   4.022             0.000121 ***
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## Residual standard error: 43.67 on 89 degrees of freedom
##   (28 observations deleted due to missingness)
## Multiple R-squared:  0.1742, Adjusted R-squared:  0.1556
## F-statistic: 9.385 on 2 and 89 DF,  p-value: 0.0002003
```

8. Use a Tukey test to make all the pairwise comparisons among the treatments.

```
library(agricolae)
HSD.test(apple_mod, "spacing", console = TRUE)
```

```
##
## Study: apple_mod ~ "spacing"
##
## HSD Test for yield
##
## Mean Square Error:  1907.304
##
## spacing,  means
##
##       yield      std  r  Min   Max
## 10 156.4893 45.60411 28 64.1 248.6
## 14 164.6724 50.41401 29 72.3 282.3
## 6  120.5657 35.32755 35 66.4 185.5
##
## Alpha: 0.05 ; DF Error: 89
## Critical Value of Studentized Range: 3.370849
##
## Groups according to probability of means differences and alpha level( 0.05 )
##
## Treatments with the same letter are not significantly different.
##
##       yield groups
## 14 164.6724      a
## 10 156.4893      a
## 6  120.5657      b
```

9. Write a few sentences that summarise your findings. What biological processes do you think drive the effects that you have detected?

There was a significant effect of spacing on apple yields (Figure XX, ANOVA: $F = 9.385$, d.f. $= 2$ and 89, $p = 0.0002$).

Then: *The pairwise comparisons in the ANOVA model showed that means of the 6 and 10 foot spacing treatment were significantly different (t= 3.244, p = 0.0017), as were those of 6 and 14 (t=4.022, p = 0.0001), but the 10 foot - 14 foot comparison showed no significant difference (t= 0.707, p= 0.4813)*[1].

Or, more simply: *The 6-10ft and 6-14ft comparisons showed significant differences (Tukey HSD: p<0.05), but the 10-14ft comparison showed no significant difference (Tukey HSD: p>0.05)*

10. Optional. Instead of using a Tukey test, use the alternative "relevel" approach to make the missing comparison.

```
apples2 <- apples %>%
  mutate(spacing = relevel(spacing, ref = "10"))
apple_mod2 <- lm(yield ~ spacing, data = apples2)
summary(apple_mod2)
```

```
##
## Call:
## lm(formula = yield ~ spacing, data = apples2)
##
## Residuals:
##     Min      1Q  Median      3Q     Max
## -92.389 -30.577  -3.516  33.192 117.628
##
## Coefficients:
##             Estimate Std. Error t value             Pr(>|t|)
## (Intercept)  156.489      8.253  18.961 < 0.0000000000000002 ***
## spacing6     -35.924     11.073  -3.244              0.00166 **
## spacing14      8.183     11.571   0.707              0.48128
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## Residual standard error: 43.67 on 89 degrees of freedom
##   (28 observations deleted due to missingness)
## Multiple R-squared:  0.1742, Adjusted R-squared:  0.1556
## F-statistic: 9.385 on 2 and 89 DF,  p-value: 0.0002003
```

If you get this far, try using the ANOVA approach on one of the previous t-test examples (remember that ANOVA can be used when your single explanatory variable has TWO or more levels). You should notice that the results are the same whether you use the `t.test` function or the ANOVA approach with `lm`.

[1] These values come from the `summary` tables for the ANOVA model, and the releveled ANOVA model

23.10 Chirping crickets

1. Import the data

```
chirps <- read.csv("CourseData/chirps.csv")
```

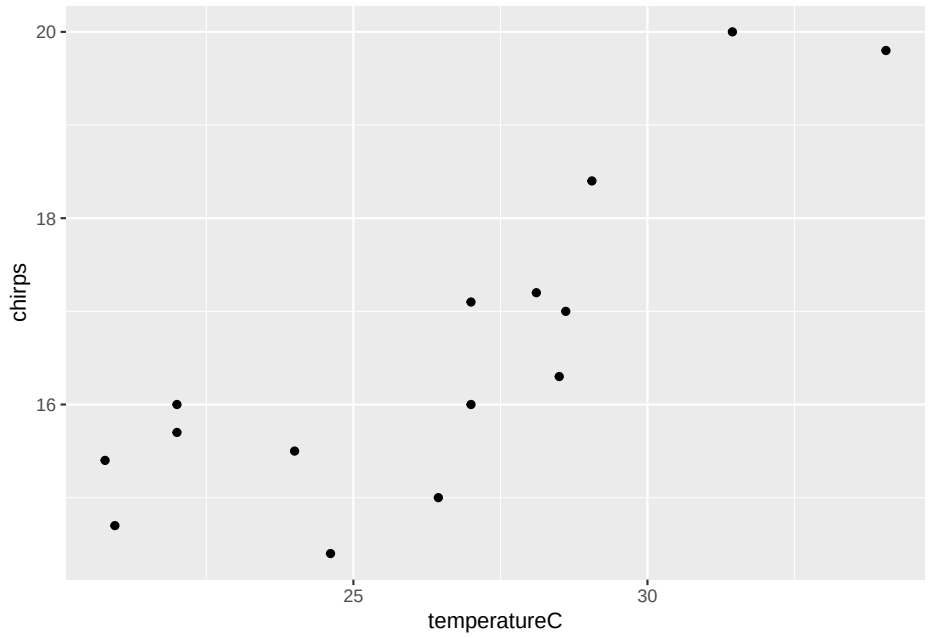
2. Use `mutate` to convert Fahrenheit to Celsius (Google it)

```
chirps <- chirps %>%
  mutate(temperatureC = (temperature - 32) * (5 / 9))
head(chirps)
```

```
##   chirps temperature temperatureC
## 1   20.0        88.6     31.44444
## 2   16.0        71.6     22.00000
## 3   19.8        93.3     34.05556
## 4   18.4        84.3     29.05556
## 5   17.1        80.6     27.00000
## 6   15.5        75.2     24.00000
```

3. Plot the data

```
(A <- ggplot(chirps, aes(x = temperatureC, y = chirps)) +
  geom_point())
```

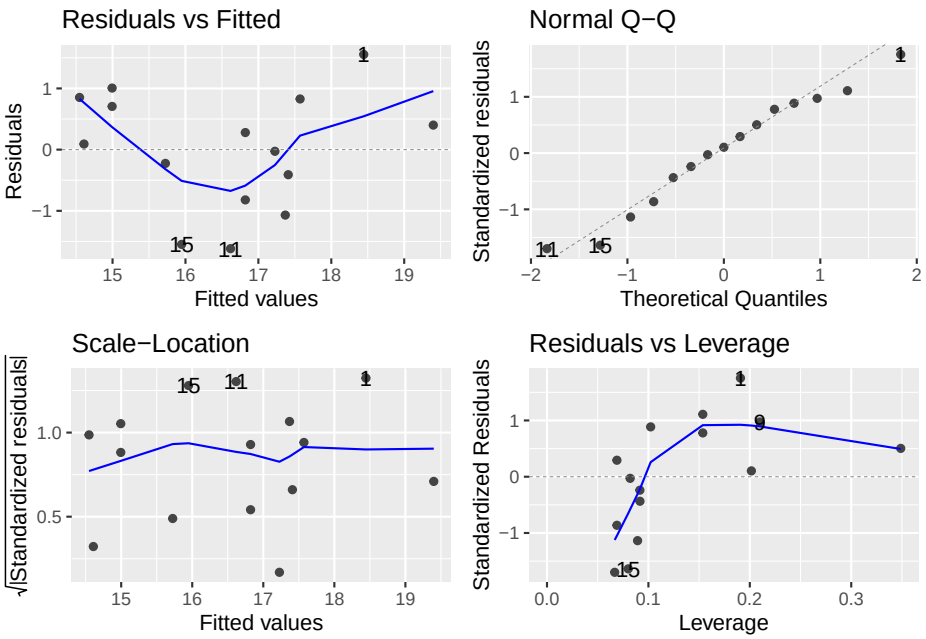


4. Fit a linear regression model with `lm`

```
chirp_mod <- lm(chirps ~ temperatureC, data = chirps)
```

5. Look at diagnostic plots to evaluate the model

```
library(ggfortify)
autoplot(chirp_mod)
```



6. Use `anova` to figure out if the effect of temperature is statistically significant.

```
anova(chirp_mod)
```

```
## Analysis of Variance Table
##
## Response: chirps
##              Df Sum Sq Mean Sq F value    Pr(>F)
## temperatureC  1 28.435 28.4348  29.248 0.0001195 ***
## Residuals    13 12.639  0.9722
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

7. Use `summary` to obtain information about the coefficients and R^2-value.

```
summary(chirp_mod)
```

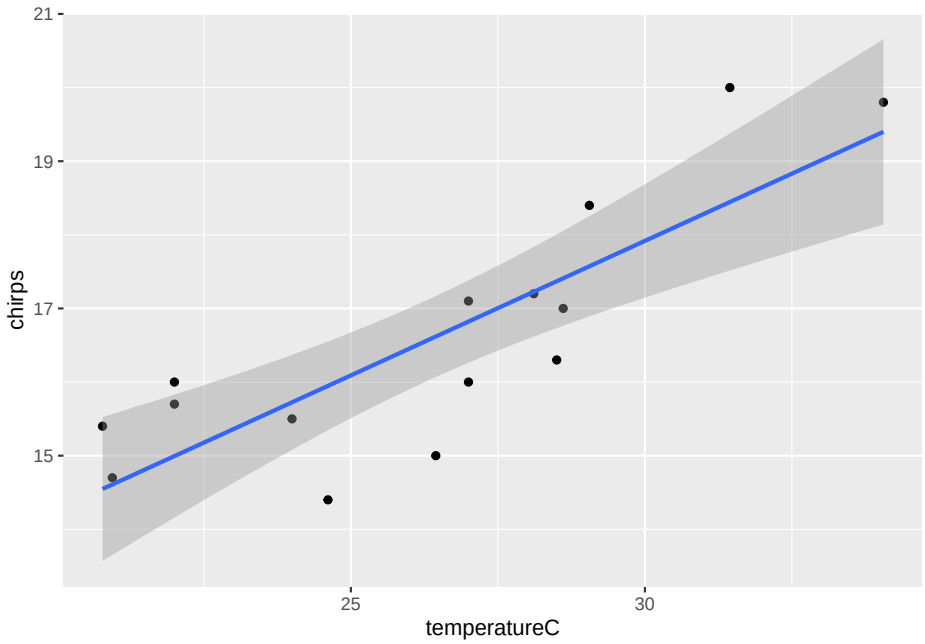
```
##
## Call:
## lm(formula = chirps ~ temperatureC, data = chirps)
##
## Residuals:
##     Min      1Q  Median      3Q     Max
## -1.6181 -0.6154  0.0916  0.7669  1.5549
##
## Coefficients:
##              Estimate Std. Error t value Pr(>|t|)
## (Intercept)   6.95531    1.79534   3.874 0.001918 **
## temperatureC  0.36540    0.06756   5.408 0.000119 ***
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## Residual standard error: 0.986 on 13 degrees of freedom
## Multiple R-squared:  0.6923, Adjusted R-squared:  0.6686
## F-statistic: 29.25 on 1 and 13 DF,  p-value: 0.0001195
```

8. Summarise the model in words.

There is a statistically significant association between temperature and chirp frequency ($F = 29.2482$, d.f. $= 1,13$, $p < 0.001$) The equation of the fitted model is: Chirp Freq $= 6.96(\pm\ 1.80) \times$ Temp $+ 0.37(\pm\ 0.07)$. The model explains 69% of the variation in chirp frequency ($R^2 = 0.692$).

9. Add model fit line to the plot.

```
A + geom_smooth(method = "lm")
```



10. Can I use cricket chirp frequency as a kind of thermometer?

Yes, using the equation from the model

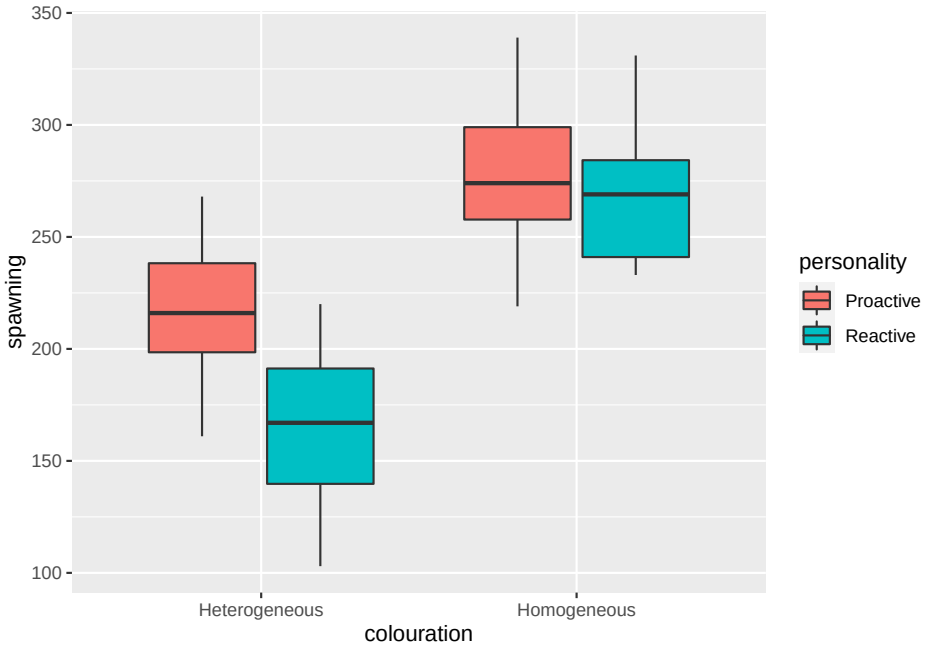
23.11 Fish behaviour

1. Import the data set, `fishPersonality.csv`

```
fishPersonality <- read.csv("CourseData/fishPersonality.csv")
```

2. Plot the data (e.g. as a box plot)

```
ggplot(fishPersonality, aes(
  x = colouration, y = spawning,
  fill = personality
)) +
  geom_boxplot()
```



3. Fit an ANOVA model using `lm`.

```
mod_A <- lm(spawning ~ personality + colouration +
  personality:colouration, data = fishPersonality)
```

4. Look at diagnostic plots from this model (`autoplot`)
5. Use `anova` to get an Analysis of Variance summary table, and interpret the results.

```
anova(mod_A)
```

```
## Analysis of Variance Table
##
## Response: spawning
##                          Df Sum Sq Mean Sq F value       Pr(>F)
## personality               1   7781    7781  5.5032      0.02629 *
## colouration               1  55862   55862 39.5074 0.0000008519 ***
## personality:colouration   1   4118    4118  2.9123      0.09898 .
## Residuals                28  39591    1414
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

6. Get the coefficient summary (`summary`) and interpret the output.

```
summary(mod_A)
```

```
##
## Call:
## lm(formula = spawning ~ personality + colouration + personality:colouration,
##     data = fishPersonality)
##
## Residuals:
##     Min      1Q  Median      3Q     Max
## -61.625 -23.344  -5.375  26.313  60.125
##
## Coefficients:
##                                           Estimate Std. Error t value
## (Intercept)                                 218.50      13.29  16.435
## personalityReactive                         -53.88      18.80  -2.865
## colourationHomogeneous                       60.87      18.80   3.238
## personalityReactive:colourationHomogeneous   45.38      26.59   1.707
##                                                    Pr(>|t|)
## (Intercept)                                0.00000000000000065 ***
## personalityReactive                                0.00781 **
## colourationHomogeneous                             0.00309 **
## personalityReactive:colourationHomogeneous         0.09898 .
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## Residual standard error: 37.6 on 28 degrees of freedom
## Multiple R-squared:  0.6312, Adjusted R-squared:  0.5917
## F-statistic: 15.97 on 3 and 28 DF,  p-value: 0.000003021
```

7. Do post-hoc Tukey tests (e.g. using `HSD.test` from the `agricolae` package). Interpret the results.

```
library(agricolae)
HSD.test(mod_A, c("personality", "colouration"), console = TRUE)
```

```
##
## Study: mod_A ~ c("personality", "colouration")
##
## HSD Test for spawning
##
## Mean Square Error:  1413.951
##
## personality:colouration,  means
##
##                           spawning      std r Min Max
## Proactive:Heterogeneous  218.500 34.92850 8 161 268
```

```
## Proactive:Homogeneous    279.375 40.40133 8 219 339
## Reactive:Heterogeneous   164.625 39.86383 8 103 220
## Reactive:Homogeneous     270.875 34.84840 8 233 331
##
## Alpha: 0.05 ; DF Error: 28
## Critical Value of Studentized Range: 3.861244
##
## Minimun Significant Difference: 51.33332
##
## Treatments with the same letter are not significantly different.
##
##                          spawning groups
## Proactive:Homogeneous    279.375      a
## Reactive:Homogeneous     270.875      a
## Proactive:Heterogeneous  218.500      b
## Reactive:Heterogeneous   164.625      c
```

8. Sum up your findings with reference to the initial research questions.

- Homogeneous coloured fish seem to do better than heterogeneous ones overall (from the plot)
- The anova table shows that personality, colouration and the interaction between them are all important variables ($p < 0.05$); Personality seems to be more important than colouration overall (based on p-values)
- The Tukey test table shows that - (1) Personality is associated with spawning, but only for heterogeneous coloured fish. (2) In the heterogeneous coloured fish the proactive fish spawn significantly more than the reactive ones. (3) In the homogeneous coloured fish there is no significant difference between the personalities.

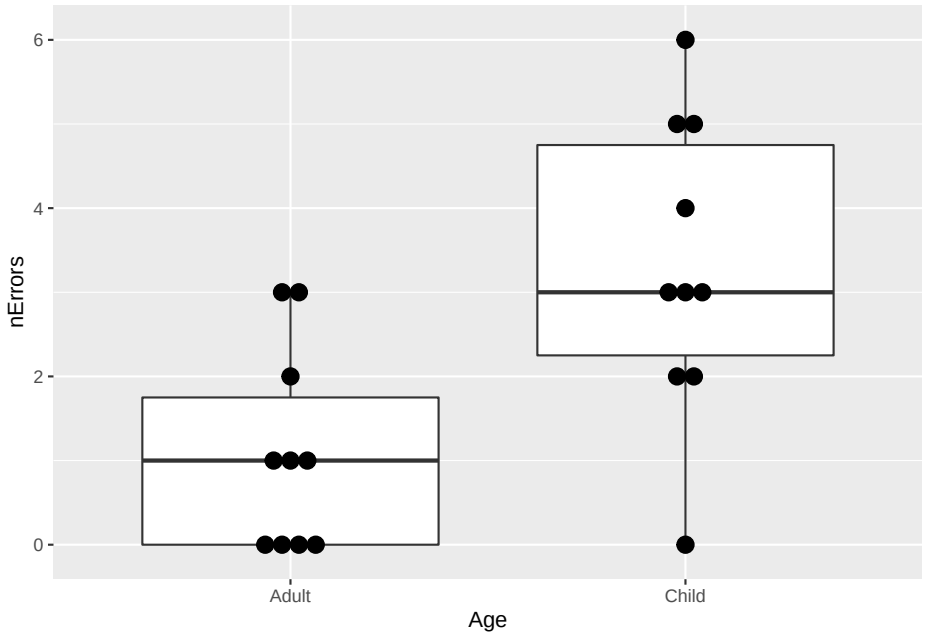
23.12 Maze runner

1. Import the data and graph it (`geom_boxplot`). Try adding the points to the `ggplot` using the new (to you) function `geom_dotplot(binaxis = "y", stackdir = "center")`.

```
maze <- read.csv("CourseData/maze.csv", stringsAsFactors = TRUE)
head(maze)
```

```
##     Age nErrors
## 1 Child       2
## 2 Child       4
## 3 Child       2
## 4 Child       5
## 5 Child       6
## 6 Child       0
```

```
(A <- ggplot(maze, aes(x = Age, y = nErrors)) +
  geom_boxplot() +
  geom_dotplot(binaxis = "y", stackdir = "center"))
```

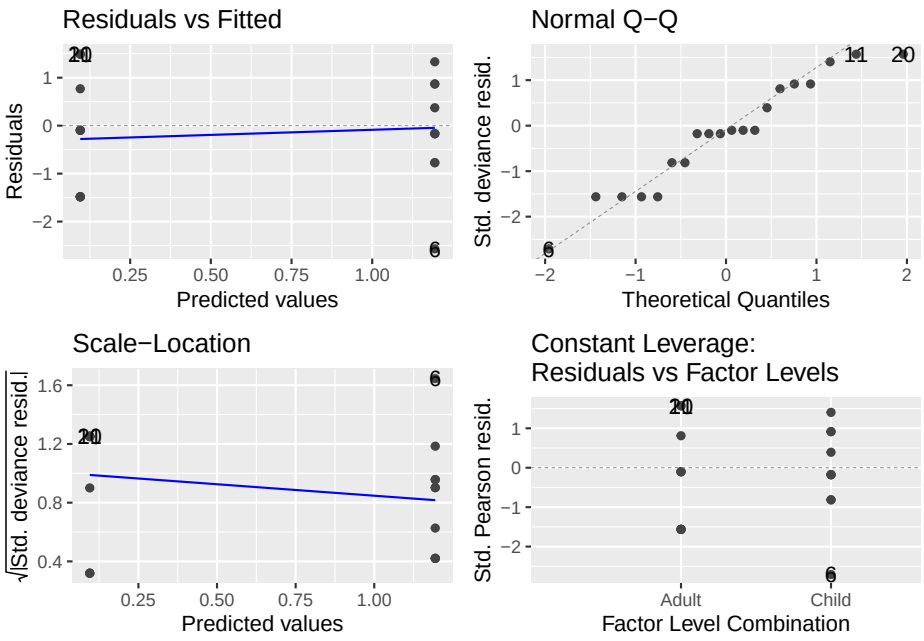


2. Fit an appropriate GLM.

```
mod_A <- glm(nErrors ~ Age, data = maze, family = poisson)
```

3. Examine the diagnostic plots of the model (`autoplot`).

```
library(ggfortify)
autoplot(mod_A)
```



4. Get the analysis of variance (deviance) table (`anova`). What does this tell you?

```
anova(mod_A, test = "Chi")
```

```
## Analysis of Deviance Table
##
## Model: poisson, link: log
##
## Response: nErrors
##
## Terms added sequentially (first to last)
##
##
##      Df Deviance Resid. Df Resid. Dev  Pr(>Chi)
## NULL                    19     36.672
## Age   1   11.511        18     25.161 0.0006917 ***
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

5. Obtain the `summary` table. What does this tell you?

```
summary(mod_A)
```

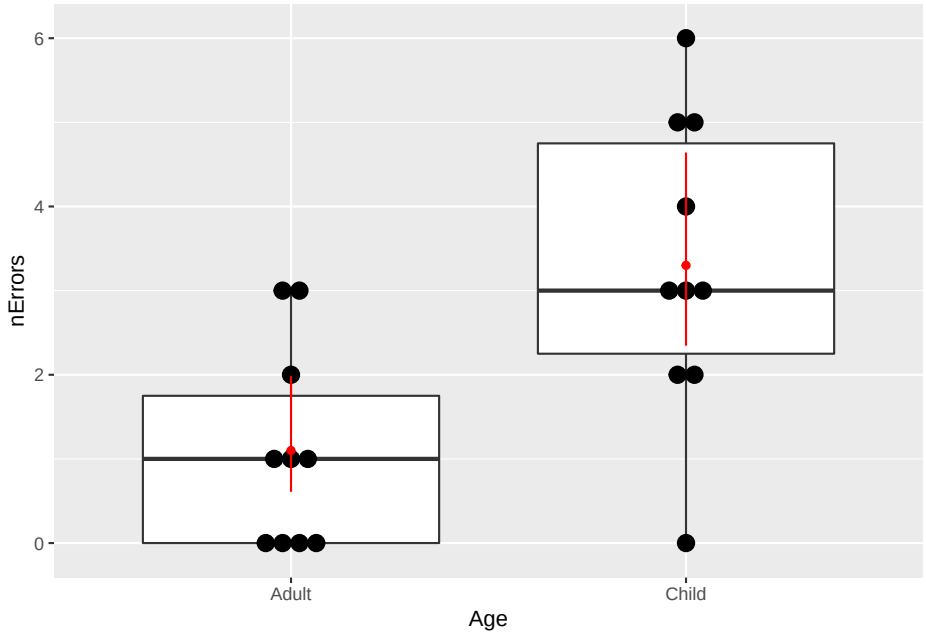
```
##
```

```
## Call:
## glm(formula = nErrors ~ Age, family = poisson, data = maze)
##
## Deviance Residuals:
##     Min       1Q   Median       3Q      Max
## -2.5690  -0.9503  -0.1323   0.7940   1.4899
##
## Coefficients:
##             Estimate Std. Error z value Pr(>|z|)
## (Intercept)  0.09531    0.30151   0.316   0.7519
## AgeChild     1.09861    0.34815   3.156   0.0016 **
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## (Dispersion parameter for poisson family taken to be 1)
##
##     Null deviance: 36.672  on 19  degrees of freedom
## Residual deviance: 25.161  on 18  degrees of freedom
## AIC: 71.845
##
## Number of Fisher Scoring iterations: 5
```

6. Use the coefficient information in the `summary` table to get the model predictions for average number of mistakes (plus/minus 95% Confidence interval). Remember that (i) the model summary is on the scale of the linear predictor, and (ii) the 95% CI can be calculated as 1.96 times the standard error. You can do these calculations "by hand", or using the `predict` function. Ask for help if you get stuck.

```
newData <- data.frame(Age = c("Child", "Adult"))
pv <- predict(mod_A, newData, se.fit = TRUE)

newData <- newData %>%
  mutate(nErrors_LP = pv$fit) %>%
  mutate(lowerCI_LP = pv$fit - 1.96 * pv$se.fit) %>%
  mutate(upperCI_LP = pv$fit + 1.96 * pv$se.fit)

inverseFunction <- family(mod_A)$linkinv

newData <- newData %>%
  mutate(nErrors = inverseFunction(nErrors_LP)) %>%
  mutate(lowerCI = inverseFunction(lowerCI_LP)) %>%
  mutate(upperCI = inverseFunction(upperCI_LP))


A + geom_point(
  data = newData, aes(x = Age, y = nErrors),
  colour = "red"
```

```
) +
  geom_segment(data = newData, aes(
    x = Age, xend = Age,
    y = lowerCI, yend = upperCI
  ), colour = "red")
```



23.13 Snails on the move

Simulate a t-test-based analysis in R to figure out what sample size would result in 80% power.

```
sampleSize <- 40 # vary this until power > 0.8
result <- replicate(
  1000,
  t.test(
    rnorm(sampleSize, mean = 7.4, sd = 2.76),
    rnorm(sampleSize, mean = 7.4 * 1.25, sd = 2.76)
  )$p.value
)
sum(result < 0.05) / 1000
```

```
## [1] 0.83
```

You should find that you need a sample size of about 40 per group.

23.14 Mouse lemur strength

What difference in strength could you reliably detect (with power >80%) with these sample sizes?

```
diff <- 185 # Vary this to give a difference to 2nd group t-test
result <- replicate(
  1000,
  t.test(
    rnorm(25, mean = 600, sd = 145),
    rnorm(8, mean = 600 - diff, sd = 145)
  )$p.value
)
sum(result < 0.05) / 1000
```

```
## [1] 0.812
```

You should find that you could detect a difference of about 185g or a mean for old animals of 415g which is 70% of the young animals i.e. a 30% reduction.