Biology 305: Biostatistics

Dr. Jacob C. Cooper & Dr. Melissa Wuellner Invalid Date

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Preface

Welcome to Biology 105 at the University of Nebraska at Kearney! Material in this class was designed by Dr. Melissa Wuellner and adapted by Dr. Jacob C. Cooper for use in R.

In this class, you will learn:

- 1. The basics of study design, the importance of understanding your research situation before embarking on a full study, and practice creating research frameworks based on different scenarios.
- 2. The basics of data analysis, including understanding what kind of variables are being collected, why understanding variable types are important, and basic tests to understand univariate distributions.
- 3. Basic multivariate statistics, including ANOVA, correlation, and regression, for comparing multiple different groups.
- 4. The basics of coding and working in R for performing statistical analyses.

This site will help you navigate different homework assignments to perform the necessary R tests. Furthermore, this GitHub repository contains all of the homework dataframes, so you will not have to manually enter assignments if you use R to complete your assignments.

Welcome to class!

Dr. Jacob C. Cooper, BHS 321

1 Intro to R

In this class, we will be using R to perform statistical analyses. R is a free software program designed for use in a myriad of statistical and computational scenarios. It can handle extremely large datasets, can handle spatial data, and has wrappers for compatibility with Python, Bash, and other programs (even Java!).

2 Setup

First, we need to download R onto your machine. We are also going to download RStudio to assist with creating R scripts and documents.

2.1 Installing *R*

First, navigate to the R download and install page. Download the appropriate version for your operating system (Windows, Mac, or Linux). **Note** that coding will be formatted slightly different for Windows than for other operating systems.

Follow the installation steps for R, and verify that the installation was successful by searching for R on your machine. You should be presented with a coding window that looks like the following:

```
Copyright (C) 2024 The R Foundation for Statistical Computing Platform: aarch64-apple-darwin20

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

Natural language support but running in an English locale

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

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

R version 4.4.1 (2024-06-14) -- "Race for Your Life"

If that screen appears, congratulations! R is properly installed. If the install was not successful, please talk to Dr. Cooper and check with your classmates as well.

2.2 Installing RStudio

RStudio is a GUI (graphics user interface) that helps make R easier to use. Furthermore, it allows you to create documents in R, including websites (such as this one), PDFs, and even presentations. This can greatly streamline the research pipeline and help you publish your results and associated code in a quick and efficient fashion.

Head over the the *RStudio* download website and download "*RStudio* Desktop", which is free. Be sure to pick the correct version for your machine.

Open RStudio on your machine. You should be presented with something like the following:

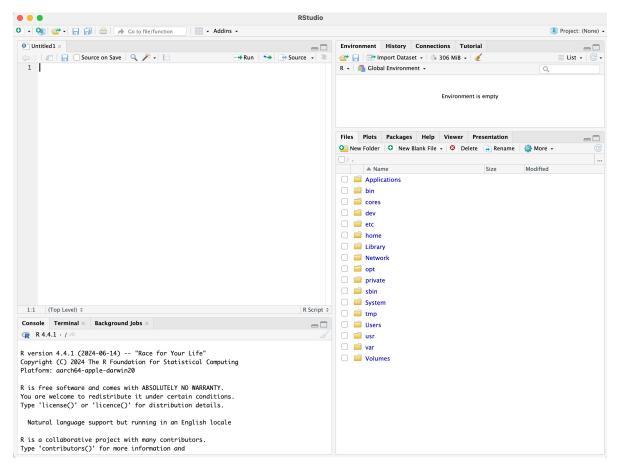


Figure 2.1: *RStudio* start window. Note that the screen is split into four different quadrants. Top left: *R* documents; bottom left: *R* program; top right: environment window; bottom right: plots, help, and directories.

In RStudio, the top left window is always going to be our coding window. This is where we will type all of our code and create our documents. In the bottom left we will see R executing the code. This will show what the computer is "thinking" and will help us spot any potential issues. The top right window is the "environment", which shows what variables and datasets are stored within the computers' memory. (It can also show some other things, but we aren't concerned with that at this point). The bottom right window is the "display" window. This is where plots and help windows will appear if they don't appear in the document (top left) window itself.

Now, we will create our first R document!

3 Creating an RMarkdown document

3.1 Setup

In this class, we will be creating assignments in what is called RMarkdown. This is a rich-text version of R that allows us to create documents with the code embedded. In RStudio, click the "+" button in the far top left to open the New Document menu. Scroll down this list and click on R Markdown.

A screen such as this will appear:

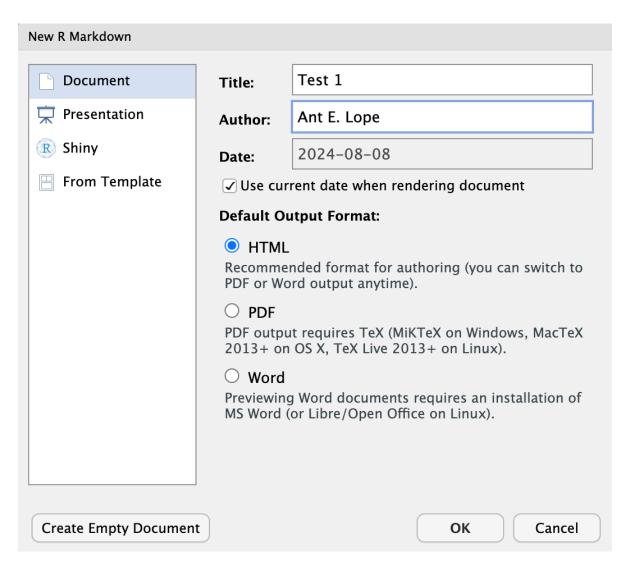


Figure 3.1: A new file window for an *RMarkdown* file.

After entering a title and your name and selecting document in the left hand menu, click OK.

```
title: "Test 1"
author: "Ant E. Lope"
date: "`r Sys.Date()`"
output: html_document
---

```{r setup, include=FALSE}
knitr::opts_chunk$set(echo = TRUE)

R Markdown

This is an R Markdown document. Markdown is a simple formatting syntax for authoring
HTML, PDF, and MS Word documents. For more details on using R Markdown see
<http://rmarkdown.rstudio.com>.

When you click the **Knit** button a document will be generated that includes both
content as well as the output of any embedded R code chunks within the document. You can
embed an R code chunk like this:

```{r cars}
summary(cars)

```
```

Figure 3.2: An example of a markdown script.

In the image above, we can see what a "default" *RMarkdown* script looks like after creating the file. At the top of the document, between all of the dashes, we have the yaml header that tells R what kind of document will be created, who the author is, and tells it to use today's date. In this class, we will be saving documents as html as they are the easiest documents to create and save. These documents will include all of your code, text, and even any plots you may create!

Plain text in the document will be rendered as plain text in the document. (I.e., whatever you type normally will become "normal text" in the finished document). Lines preceded with # will become headers, with ## being a second level header and ### being a third level header, etc. Words can also be made italic by putting an asterisk on each side of the word (\*italic\*) and bold by putting two asterisks on each side (\*\*bold\*\*). URLs are also supported, with <> on each side of a URL making it clickable, and words being hyperlinked by typing [words to show] (target URL).

We also have code "chunks" that are shown above. A code chunk can be manually typed out or inserted by pressing CTRL + ALT + I (Windows, Linux) or COMMAND + OPTION + I (Mac). Everything inside a "code chunk" will be read as R code and executed as such. Note that you can have additional commands in the R chunks, but we won't cover that for now.

## 3.2 Using code chunks

In your computer, erase all information except for the yaml header between the dashes on your computer. Save your file in a folder where you want your assignment to be located. It is important you do this step up front as the computer will sometimes save in random places if you don't specify a file location at the beginning. Don't forget to save your work frequently!

```
This is a test of the *RMarkdown* code.

```{r}
print("Hello world!")

```
```

Figure 3.3: Text to type in your *Rmarkdown* document.

After typing this into the document, hit knit near the top of the upper left window. R will now create an HTML document that should look like this:

## Test 1

### Ant E. Lope 2024-08-07

This is a test of the RMarkdown code.

```
print("Hello world!")

[1] "Hello world!"
```

Figure 3.4: The output from the above code knitted into a document.

We can see now that the HTML document has the title of the document, the author's name, the date on which the code was run, and a greyed-out box with color coded R code followed by the output. Let's try something a little more complex. Create a new code chunk and type the following:

```
x <- 1:10
```

This will create a variable in R, x, that is sequentially each whole number between 1 and 10. We can see this by highlighting or typing only the letter x and running that line of code by clicking CTRL + ENTER (Windows / Linux) or COMMAND + ENTER (Mac).

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

If you look at the top right window, you will also see the value x in the environment defined as int [1:10] 1 2 3 4 5 6 7 8 9 10. This indicates that x is integer data spanning ten positions numbered 1 to 10. Since the vector is small, it displays every number in the sequence.

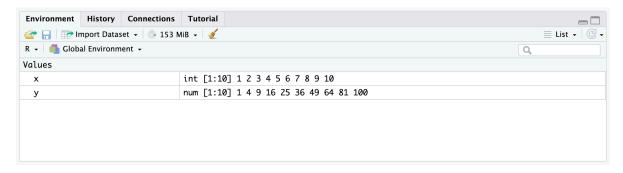


Figure 3.5: *RStudio* environment window showing saved objects. These are in the computer's memory.

Let's create another vector y that is the squared values of x, such that  $y = x^2$ . We can raise values to an exponent by using  $\hat{}$ .

```
y <- x²
y
```

#### [1] 1 4 9 16 25 36 49 64 81 100

Now we have the value y in the environment that is the square of the values of x. This is a numeric vector of 10 values numbered 1 to 10 where each value corresponds to a square of the x value. We can raise things to any value however, including  $x^x$ !

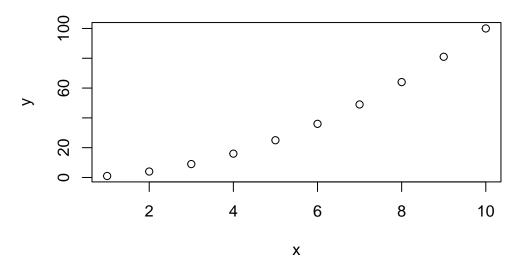
| $x^x$ |        |          |           |             |      |       |
|-------|--------|----------|-----------|-------------|------|-------|
|       |        |          |           |             |      |       |
| [1]   | 1      | 4        | 27        | 256         | 3125 | 46656 |
| [7]   | 823543 | 16777216 | 387420489 | 10000000000 |      |       |

As we can see, since I didn't "store" this value as a variable in R using  $\leftarrow$ , the value is not in the environment.

## 3.3 Plotting

Now, let's try creating a plot. This is easy in R, as we just use the command plot.

$$plot(x = x, y = y)$$



By specifying the y and x components in plot, we can quickly generate a point plot. We can alter the visual parameters of this plot using a few different commands. I will outline these below with inline notes. Inline notes in the code can be made by using a # symbol before them, which basically tells R to ignore everything after the #. For example:

```
print("Test")
```

#### [1] "Test"

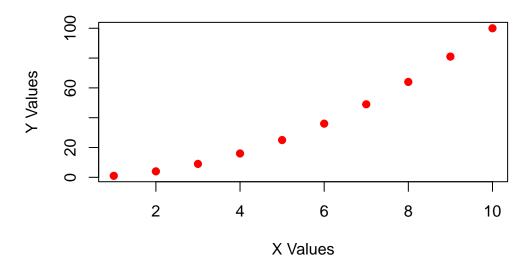
```
print("Test 2")
```

This prints the word Test, but doesn't print Test 2.

Now let's make the plot with some new visual parameters.

```
plot(x = x, # specify x values
 y = y, # specify y values
 ylab = "Y Values", # specify Y label
 xlab = "X Values", # specify X label
 main = "Plot Title", # specify main title
 pch = 19, # adjust point style
 col = "red") # make points red
```

### **Plot Title**



## 3.4 Tab complete

RStudio allows for "tab-completing" while typing code. Tab-completing is a way of typing the first part of a command, variable name, or file name and hitting "tab" to show all options with that spelling. You should use tab completing because it:

- reduces spelling mistakes
- reduces filepath mistakes
- increases the speed at which you code
- provides help with specific functions

## 3.5 Help

At any point in R, you can look up "help" for a specific function by typing ?functionname. Try this on your computer with the following:

?mean

## 4 Working with data

Throughout this course, we are going to have to work with datasets that are from our book or other sources. Here, we are going to work through an example dataset. First, we need to install *libraries*. A *library* is a collated, pre-existing batch of code that is designed to assist with data analysis or to perform specific functions. These *libraries* make life a lot easier, and create short commands for completing relatively complex tasks.

In this class, there are two libraries that you will need *almost every week*! First, we need to install the libraries. The main libraries we need for this class are:

- 1. curls: this package allows us to download things from URLs. We will be using this to download data files. Otherwise, you will have to enter all data by hand!
- 2. tidyverse: this package is actually a group of packages designed to help with data analysis, management, and visualization.

```
run this code the first time ONLY
does not need to be run every time you use R!

curl allows for internet downloads
install.packages("curl")

tidyverse has a bunch of packages in it!
great for data manipulation
install.packages("tidyverse")

if you ever need to update:
leaving brackets open means "update everything"
update.packages()
```

After packages are installed, we will need to load them into our R environment. While we only need to install.packages once on our machine, we need to load libraries every time we restart the program!

```
library(curl)
```

Using libcurl 8.7.1 with LibreSSL/3.3.6

#### library(tidyverse)

```
-- Attaching core tidyverse packages ---
 ---- tidyverse 2.0.0 --
v dplyr
 1.1.4
 2.1.5
 v readr
v forcats
 1.0.0
 v stringr
 1.5.1
v ggplot2
 3.5.1
 v tibble
 3.2.1
v lubridate 1.9.3
 v tidyr
 1.3.1
v purrr
 1.0.2
-- Conflicts -----
 -----ctidyverse_conflicts() --
x dplyr::filter()
 masks stats::filter()
x dplyr::lag()
 masks stats::lag()
x readr::parse_date() masks curl::parse_date()
i Use the conflicted package (http://conflicted.r-lib.org/) to force all conflicts to become
```

You should an output like the above. What this means is:

- 1. The core packages that comprise the tidyverse loaded successfully, and version numbers for each are shown.
- 2. The conflicts basically means that certain commands will not work as they used to because R has "re-learned" a particular word.

To clarify the conflicts, pretend that you can only know one definition of a word at a time. You may know the word "cola" as a type of soda pop or as a drink in general. However, in Spanish, "cola" refers to a line or a tail. While we can learn both of these definitions and know which one is which because of context, a computer can't do that. In R, we would then have to specify which "cola" we are refferring to. We do this by listing the package before the command; in this case, english::cola would mean a soda pop and spanish::cola would refer to a line or tail. If we just type cola, the computer will assume one of these definitions but not even consider the other.

We won't have to deal with conflicts much in this class, and I'll warn you (or help you) if there is a conflict.

## 4.1 Downloading data

Now, we need to download our first data set. These datasets are stored on GitHub. We are going to be looking at data from Dr. Cooper's dissertation concerning Afrotropical bird distributions (Cooper 2021). This website is in the data folder on this websites' GitHub page, accessible here.

Alternatively, we can use the operator %% to simplify this process. %%% means "take whatever you got from the previous step and pipe it into the next step". So, the following does the exact same thing:

```
ranges <- curl("https://raw.githubusercontent.com/jacobccooper/biol105_unk/main/datasets/lac-
read_csv()
```

Rows: 12 Columns: 10

Using the %>% is preferred as you can better set up a workflow and because it more closely mimics other coding languages, such as bash.

Let's view the data to see if it worked. We can use the command head to view the first few rows:

#### head(ranges)

```
A tibble: 6 x 10
 species
 combined_current_km2 consensus_km2 bioclim_current_km2
 <chr>
 <dbl>
 <dbl>
 <dbl>
 25209.
1 Batis diops
 6694.
 19241.
2 Chamaetylas_poliophrys
 68171.
 1106.
 68158.
3 Cinnyris_regius
 60939.
 13305.
 53627.
4 Cossypha_archeri
 27021.
 6409.
 11798.
5 Cyanomitra alinae
 78680.
 34320.
 63381.
6 Graueria_vittata
 8770.
 861.
 8301.
i 6 more variables: `2050_combined_km2` <dbl>, `2050_consensus_km2` <dbl>,
 `2070_combined_km2` <dbl>, `2070_consensus_km2` <dbl>,
#
 alltime_consensus_km2 <dbl>, past_stable_km2 <dbl>
```

We can perform a lot of summary statistics in R. Some of these we can view for multiple columns at once using summary.

#### summary(ranges)

```
species
 combined_current_km2 consensus_km2
 bioclim_current_km2
Length:12
 Min.
 : 8770
 Min.
 : 861.3
 Min.
 : 3749
Class :character
 1st Qu.: 24800
 1st Qu.: 4186.2
 1st Qu.: 10924
Mode :character
 Median: 43654
 Median : 7778.1
 Median : 31455
 Mean
 : 68052
 Mean
 :18161.8
 Mean
 : 42457
 3rd Qu.: 70798
 3rd Qu.:18558.7
 3rd Qu.: 62835
 Max.
 :232377
 Max.
 :79306.6
 Max.
 :148753
2050_combined_km2 2050_consensus_km2 2070_combined_km2
 2070_consensus_km2
 : 1832
 0.0
 550.3
 0.0
Min.
 Min.
 Min.
 Min.
1st Qu.:
 6562
 1st Qu.: 589.5
 1st Qu.: 6583.8
 1st Qu.: 311.4
Median : 26057
 Median: 6821.9
 Median : 24281.7
 Median: 2714.6
Mean
 : 33247
 :14418.4
 Mean
 : 31811.0
 : 8250.5
 Mean
 Mean
3rd Qu.: 40460
 3rd Qu.:18577.1
 3rd Qu.: 38468.9
 3rd Qu.:10034.4
Max.
 :132487
 Max.
 :79236.2
 Max.
 :129591.0
 Max.
 :53291.8
alltime consensus km2 past stable km2
 0.0
 Min.
 :
1st Qu.: 790.9
 1st Qu.:
 0.0
Median: 8216.8
 Median:
 0.0
 :15723.3
 : 127.3
Mean
 Mean
3rd Qu.:19675.0
 3rd Qu.:
 0.0
Max.
 :82310.5
 Max.
 :1434.8
```

As seen above, we now have information for the following statistics for each variable:

- Min = minimum
- 1st Qu. = 1st quartile
- Median = middle of the dataset
- Mean = average of the dataset
- 3rd Qu. = 3rd quartile
- Max. = maximum

We can also calculate some of these statistics manually to see if we are doing everything correctly. It is easiest to do this by using predefined functions in R (code others have written to perform a particular task) or to create our own functions in R. We will do both to determine the average of combined\_current\_km2.

### 4.2 Subsetting data

First, we need to select only the column of interest. In R, we have two ways of subsetting data to get a particular column.

- var[rows,cols] is a way to look at a particular object (var in this case) and choose a specific combination of row number and column number (col). This is great if you know a specific index, but it is better to use a specific name.
- var[rows,"cols"] is a way to do the above but by using a specific column name, like combined\_current\_km2.
- var\$colname is a way to call the specific column name directly from the dataset.

```
using R functions
ranges$combined_current_km2
```

```
[1] 25209.4 68171.2 60939.2 27021.3 78679.9 8769.9 232377.2 17401.4 [9] 51853.5 35455.1 23570.3 187179.1
```

As shown above, calling the specific column name with \$ allows us to see only the data of interest. We can also save these data as an object.

```
current_combined <- ranges$combined_current_km2
current_combined</pre>
```

```
[1] 25209.4 68171.2 60939.2 27021.3 78679.9 8769.9 232377.2 17401.4 [9] 51853.5 35455.1 23570.3 187179.1
```

Now that we have it as an object, specifically a numeric vector, we can perform whatever math operations we need to on the dataset.

```
mean(current_combined)
```

#### [1] 68052.29

Here, we can see the mean for the entire dataset. However, we should always round values to the same number of decimal points as the original data. We can do this with round.

```
round(mean(current_combined),1) # round mean to one decimal
```

#### [1] 68052.3

*Note* that the above has a nested set of commands. We can write this exact same thing as follows:

```
pipe mean through round
mean(current_combined) %>%
 round(1)
```

#### [1] 68052.3

Use the method that is easiest for you to follow!

We can also calculate the mean manually. The mean is  $\frac{\sum_{i=1}^{n} x}{n}$ , or the sum of all the values within a vector divided by the number of values in that vector.

```
create function
use curly brackets to denote function
our data goes in place of "x" when finally run
our_mean <- function(x){
 sum_x <- sum(x) # sum all values in vector
 n <- length(x) # get length of vector
 xbar <- sum_x/n # calcualte mean
 return(xbar) # return the value outside the function
}</pre>
```

Let's try it.

```
our_mean(ranges$combined_current_km2)
```

[1] 68052.29

As we can see, it works just the same as mean! We can round this as well.

```
our_mean(ranges$combined_current_km2) %>%
 round(1)
```

[1] 68052.3

# 5 Your turn!

With a partner or on your own, try to do the following:

- 1. Create an *RMarkdown document* that will save as an .html.
- 2. Load the data, as shown here, and print the summary statistics in the document.
- 3. Calculate the value of  $combined\_current\_km2$  divided by 2050\_combined\_km2 and print the results.

Let me know if you have any issues.

## **6 Descriptive Statistics**

### 6.1 Purposes of descriptive statistics

Descriptive statistics enable researchers to quickly and easily examine the "behavior" of their datasets, identifying potential errors and allowing them to observe particular trends that may be worth further analysis. Here, we will cover how to calculate descriptive statistics for multiple different datasets, culminating in an assignment covering these topics.

## 6.2 Preparing R

As with every week, we will need to load our relevant packages first. This week, we are using the following:

```
allows for internet downloading
library(curl)
```

Using libcurl 8.7.1 with LibreSSL/3.3.6

```
enables data management tools
library(tidyverse)
```

```
-- Attaching core tidyverse packages ----
 ----- tidyverse 2.0.0 --
v dplyr
 1.1.4
 2.1.5
 v readr
 1.0.0
v forcats
 v stringr
 1.5.1
v ggplot2
 3.5.1
 v tibble
 3.2.1
v lubridate 1.9.3
 v tidyr
 1.3.1
v purrr
 1.0.2
-- Conflicts -----
 ----- tidyverse_conflicts() --
x dplyr::filter()
 masks stats::filter()
x dplyr::lag()
 masks stats::lag()
x readr::parse_date() masks curl::parse_date()
i Use the conflicted package (http://conflicted.r-lib.org/) to force all conflicts to become
```

## 6.3 Downloading the data

For the example this week, we will be using the starbucks dataset, describing the number of drinks purchased during particular time periods during the day.

```
starbucks <- curl("https://raw.githubusercontent.com/jacobccooper/biol105_unk/main/datasets/
read_csv()</pre>
```

```
Rows: 9 Columns: 2
-- Column specification ------
Delimiter: ","
chr (1): Hour
dbl (1): Frap_Num

i Use `spec()` to retrieve the full column specification for this data.
```

i Specify the column types or set `show\_col\_types = FALSE` to quiet this message.

## 6.4 Descriptive statistics

Descriptive statistics are statistics that help us understand the shape and nature of the data on hand. These include really common metrics such as mean, median, and mode, as well as more nuanced metrics like quartiles that help us understand if there is any skew in the dataset. (Skew refers to a bias in the data, where more data points lie on one side of the distribution and there is a long tail of data in the other direction).

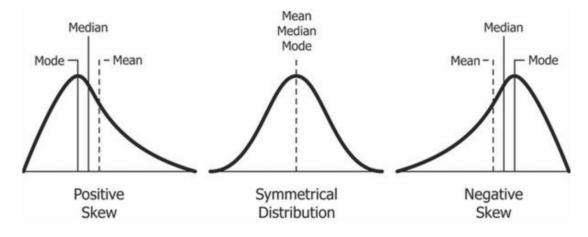


Figure 6.1: Examples of skew compared to a symmetrical, non-skewed distribution. Source: machinelearningparatodos.com

Note above that the relative position of the mean, median, and mode can be indicative of skew. Please also note that these values will rarely be exactly equal "in the real world", and thus you need to weigh differences against the entire dataset when assessing skew. There is a lot of nuance like this in statistics; it is not always an "exact" science, but sometimes involves judgment calls and assessments based on what you observe in the data.

Using the **starbucks** dataset, we can look at some of these descriptive statistics to understand what is going on.

#### 6.4.1 Notation

As a quick reminder, we use Greek lettering for *populations* and Roman lettering for samples. For example:

- $\sigma$  is a population, but s is a sample (both these variables refer to standard deviation).
- $\mu$  is a population, but  $\bar{x}$  is a sample (both of these variables refer to the *mean*).

#### 6.4.2 Mean

The mean is the "average" value within a set of data, specifically, the sum of all values divided by the length of those values:  $\frac{\sum_{i=1}^{n} x}{n}$ .

#### head(starbucks)

```
A tibble: 6 x 2
 Hour
 Frap_Num
 <chr>
 <dbl>
1 0600-0659
 2
2 0700-0759
 3
3 0800-0859
 2
4 0900-0959
 4
5 1000-1059
 8
6 1100-1159
 7
```

Here, we are specifically interested in the number of frappuccinos.

```
get vector of frappuccino number
fraps <- starbucks$Frap_Num

get mean of vector
mean(fraps)</pre>
```

#### [1] 6.222222

*Note* that the above should be rounded to a whole number, since we were given the data in whole numbers!

```
mean(fraps) %>%
 round(0)
```

#### [1] 6

We already covered calculating the average manually in our previous tutorial, but we can do that here as well:

```
sum values
divide by n, length of vector
round to 0 places
round(sum(fraps)/length(fraps),0)
```

[1] 6

#### **6.4.3** Range

The range is the difference between the largest and smallest units in a dataset. We can use the commands min and max to calculate this.

```
max(fraps) - min(fraps)
```

[1] 13

The range of our dataset is 13.

#### 6.4.4 Median

The median is also known as the 50th percentile, and is the midpoint of the data when ordered from least to greatest. If there are an even number of data points, then it is the average point between the two center points. For odd data, this is the  $\frac{n+1}{2}$ th observation. For even data, since we need to take an average, this is the  $\frac{\frac{n}{2}+(\frac{n}{2}+1)}{2}$ . You should be able to do these by hand and by using a program.

### median(fraps)

#### [1] 4

Now, to calculate by hand:

```
length(fraps)
```

#### [1] 9

We have an odd length.

```
order gets the order
order(fraps)
```

#### [1] 1 3 7 2 4 6 5 9 8

```
[] tells R which elements to put where
frap_order <- fraps[order(fraps)]
frap_order</pre>
```

#### [1] 2 2 2 3 4 7 8 13 15

```
always use parentheses
make sure the math maths right!
(length(frap_order)+1)/2
```

#### [1] 5

Which is the fifth element in the vector?

#### frap\_order[5]

### [1] 4

Now let's try it for an even numbers.

```
remove first element
even_fraps <- fraps[-1]

even_fraps_order <- even_fraps[order(even_fraps)]

even_fraps_order</pre>
```

[1] 2 2 3 4 7 8 13 15

```
median(even_fraps)
```

[1] 5.5

Now, by hand:  $\frac{n}{2} + (\frac{n}{2} + 1)$ .

```
n <- length(even_fraps_order)

get n/2 position from vector

m1 <- even_fraps_order[n/2]

get n/2+1 position

m2 <- even_fraps_order[(n/2)+1]

add these values, divide by two for "midpoint"

med <- (m1+m2)/2

med</pre>
```

[1] 5.5

As we can see, these values are equal!

#### 6.4.5 Other quartiles and quantiles

We also use the 25th percentile and the 75th percentile to understand data distributions. These are calculated similar to the above, but the bottom quartile is only  $\frac{1}{4}$  of the way between values and the 75th quartile is  $\frac{3}{4}$  of the way between values. We can use the R function quantile to calculate these.

#### quantile(frap\_order)

```
0% 25% 50% 75% 100%
2 2 4 8 15
```

We can specify a quantile as well:

```
quantile(frap_order, 0.75)
```

75% 8

We can also calculate these metrics by hand. Let's do it for the even dataset, since this is more difficult.

```
quantile(even_fraps_order)
```

```
0% 25% 50% 75% 100%
2.00 2.75 5.50 9.25 15.00
```

Note that the 25th and 75th percentiles are also between two different values. These can be calculated as a quarter and three-quarters of the way between their respective values.

```
75th percentile

n <- length(even_fraps_order)

get position
p <- 0.75*(n+1)

get lower value
round down
m1 <- even_fraps_order[trunc(p)]

get upper value
round up
m2 <- even_fraps_order[ceiling(p)]

position between</pre>
```

```
fractional portion of rank
frac <- p-trunc(p)

calculate the offset from lowest value
val <- (m2 - m1)*frac

get value
m1 + val</pre>
```

[1] 11.75

Wait... why does our value differ?

R, by default, calculates quantiles using what is called Type 7, in which the quantiles are calculated by  $p_k = \frac{k-1}{n-1}$ , where n is the length of the vector and k refers to the quantile being used. However, in our book and in this class, we use Type 6 interpretation -  $p_k = \frac{k}{n+1}$ . Let's try using Type 6:

```
quantile(even_fraps_order, type = 6)
```

```
0% 25% 50% 75% 100%
2.00 2.25 5.50 11.75 15.00
```

Now we have the same answer as we calculated by hand!

This is a classic example of how things in R (and in statistics in general!) can depend on interpretation and are not always "hard and fast" rules.

In this class, we will be using Type 6 interpretation for the quantiles - you will have to specify this in the quantile function EVERY TIME! If you do *not* specify Type 6, you will get the questions incorrect and you will get answers that do not agree with the book, with Excel, or what you calculate by hand.

#### 6.4.6 Mode

There is no default method for finding the mode in R. However, websites like Statology provide wraparound functions.

```
Statology function
define function to calculate mode
find_mode <- function(x) {
 # get unique values from vector
 u <- unique(x)
 # count number of occurrences for each value
 tab <- tabulate(match(x, u))
 # return the value with the highest count
 u[tab == max(tab)]
}
find_mode(fraps)</pre>
```

#### [1] 2

We can also do this by hand, by counting the number of occurrences of each value. This can be done in a stepwise fashion using commands in the above function.

```
unique counts
u <- unique(fraps)
u</pre>
```

#### [1] 2 3 4 8 7 15 13

```
which elements match
match(fraps,u)
```

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

```
count them
tab <- match(fraps,u) %>%
 tabulate()
tab
```

#### [1] 3 1 1 1 1 1 1

Get the highest value.

#### u[tab==max(tab)]

#### [1] 2

Notice this uses ==. This is a logical argument that means "is equal to" or "is the same as". For example:

2 == 2

#### [1] TRUE

These values are the same, so TRUE is returned.

2 == 3

#### [1] FALSE

These values are unequal, so FALSE is returned. R will read TRUE as 1 and FALSE as ZERO, such that:

sum(2==2)

[1] 1

and

sum(2==3)

#### [1] 0

This allows you to find how many arguments match your condition quickly, and even allows you to subset based on these indices as well. Keep in mind you can use greater than <, less than >, greater than or equal to <=, less than or equal to >=, is equal to ==, and is not equal to != to identify numerical relationships. Other logical arguments include:

• &: both conditions must be TRUE to match (e.g., c(10,20) & c(20,10)). Try the following as well: fraps < 10 & fraps > 3.

- &&: and, but works with single elements and allows for better parsing. Often used with if. E.g., fraps < 10 && fraps > 3. This will not work on our multi-element frap vector.
- |: or, saying at least one condition must be true. Try: fraps > 10 | fraps < 3.
- ||: or, but for a single element, like && above.
- !: not, so "not equal to" would be !=.

#### 6.4.7 Variance

When we are dealing with datasets, the variance is a measure of the total spread of the data. The variance is calculated using the following:

$$\sigma^2 = \frac{\sum (x_i - \bar{x})^2}{n - 1}$$

Essentially, this means that for every value of x, we are finding the difference between that value and the mean and squaring it, summing all of these quared differences, and dividing them by the number of samples in the dataset minus one. Let's do this for the frappuccino dataset.

#### frap\_order

[1] 2 2 2 3 4 7 8 13 15

Now to find the differences.

```
diffs <- frap_order - mean(frap_order)
diffs</pre>
```

Note that R is calculating the same thing for the entire vector! Since these are differences from the mean, they should sum to zero.

#### sum(diffs)

[1] 3.552714e-15

```
square differences
diffs_sq <- diffs^2
diffs_sq</pre>
```

```
[1] 17.8271605 17.8271605 17.8271605 10.3827160 4.9382716 0.6049383 3.1604938 [8] 45.9382716 77.0493827
```

Now we have the squared differences. We need to sum these and divide by n-1.

```
n <- length(frap_order)
var_frap <- sum(diffs_sq)/(n-1)
var_frap</pre>
```

#### [1] 24.44444

Let's check this against the built-in variance function in R.

```
var(frap_order)
```

#### [1] 24.44444

They are identical! We can check this using a logical argument.

```
var_frap == var(frap_order)
```

[1] TRUE

Seeing as this is TRUE, we calculated it correctly.

#### 6.4.8 Standard deviation

Another common measurement of spread is the standard deviation  $(\sigma)$ . As you remember from class (or may have guessed from the notation on this site), the standard deviation is just the square root of the variance.

```
sqrt(var_frap)
```

[1] 4.944132

We can test this against the built in sd function in R:

```
sqrt(var_frap) == sd(frap_order)
```

[1] TRUE

As you can see, we calculated this correctly!

#### 6.4.9 Standard error

The standard error is used to help understand the spread of data and to help estimate the accuracy of our measurements for things like the mean. The standard error is calculated thusly:

$$SE = \frac{\sigma}{\sqrt{n}}$$

There is not built in function for the standard error in excel, but we can write our own:

```
se <- function(x){
 n <- length(x) # calculate n
 s <- sd(x) # calculate standard deviation
 se_val <- s/sqrt(n)
 return(se_val)
}</pre>
```

Let's test this code.

```
se(frap_order)
```

[1] 1.648044

Our code works! And we can see exactly how the standard error is calculate. We can also adjust this code as needed for different situations, like samples.

**Remember**, the standard error is used to help reflect our *confidence* in a specific measurement (e.g., how certain we are of the mean, and what values we believe the mean falls between). We want our estimates to be as precise as possible with as little uncertainty as possible. Given this, does having more samples make our estimates more or less confident? Mathematically, what happens as our sample size *increases*?

#### 6.4.10 Coefficient of variation

The coefficient of variation, another measure of data spread and location, is calculated by the following:

$$CV = \frac{\sigma}{\mu}$$

We can write a function to calculate this in R as well.

```
cv <- function(x){
 sigma <- sd(x)
 mu <- mean(x)
 val <- sigma/mu
 return(val)
}</pre>
```

#### [1] 0.7945927

Remember that we will need to round values.

#### 6.4.11 Outliers

Outliers are any values that are outside of the 1.5 times the interquartile range. We can calculate this for our example dataset as follows:

```
lowquant <- quantile(frap_order, 0.25, type = 6) %>% as.numeric()
hiquant <- quantile(frap_order, 0.75, type = 6) %>% as.numeric()
iqr <- hiquant - lowquant
lowbound <- mean(frap_order) - (1.5*iqr)
hibound <- mean(frap_order) + (1.5*iqr)

low outliers?
select elements that match
identify using logical "which"
frap_order[which(frap_order < lowbound)]</pre>
```

numeric(0)

```
high outliers?
select elements that match
identify using logical "which"
frap_order[which(frap_order > hibound)]
```

numeric(0)

We have no outliers for this particular dataset.

### 6.5 Homework: Descriptive Statistics

Now that we've covered these basic statistics, it's your turn! For this week, you will be completing homework that involves methods from Chapter 4 in your book.

#### 6.5.1 Homework instructions

Please create an RMarkdown document that will render as an .html file. You will submit this file to show your coding and your work. Please refer to the Introduction to R for refreshers on how to create an .html document in RMarkdown. You will need to do the following for each of these datasets:

• mean

- median
- range
- interquartile range
- variance
- standard deviation
- coefficient of variation
- standard error
- whether there are any "outliers"

Please show all of your work for full credit.

#### 6.5.2 Data for homework problems

For each question, calculate the mean, median, range, interquartile range, variance, standard deviation, coefficient of variation, standard error, and whether there are any "outliers".

Please also write your own short response to the *Synthesis question* posed, which will involve thinking about the data and metrics you just analyzed.

#### 6.5.2.1 1: UNK Nebraskans

Ever year, the university keeps track of where students are from. The following are data on the number of students admitted to UNK from the state of Nebraska:

| years | nebraskans                                                                   |
|-------|------------------------------------------------------------------------------|
| 2023  | 5056                                                                         |
| 2022  | 5061                                                                         |
| 2021  | 5276                                                                         |
| 2020  | 5244                                                                         |
| 2019  | 5209                                                                         |
| 2018  | 5262                                                                         |
| 2017  | 5466                                                                         |
| 2016  | 5606                                                                         |
| 2015  | 5508                                                                         |
| 2014  | 5540                                                                         |
| 2013  | 5614                                                                         |
|       | 2023<br>2022<br>2021<br>2020<br>2019<br>2018<br>2017<br>2016<br>2015<br>2014 |

Using these data, please calculate the mean, median, range, interquartile range, variance, standard deviation, coefficient of variation, standard error, and whether there are any "outliers" for the number of UNK students from Nebraska.

**Synthesis question**: Do you think that there are any really anomalous years? Do you feel data are similar between years? *Note* we are not looking at trends through time but whether any years are outliers.

#### 6.5.2.2 2: Piracy in the Gulf of Guinea

The following is a dataset looking at oceanic conditions and other variables associated with pirate attacks within the region between 2010 and 2021 (Moura et al. 2023). Using these data, please calculate the mean, median, range, interquartile range, variance, standard deviation, coefficient of variation, standard error, and whether there are any "outliers" for distance from shore for each pirate attack (columns Distance\_from\_Coast).

```
url_file <- curl("https://figshare.com/ndownloader/files/42314604")
pirates <- url_file %>% read_csv()
```

**Synthesis question**: Do you notice any patterns in distance from shore? What may be responsible for these patterns? *Hint*: Think about what piracy entails and also what other columns are available as other variables in the above dataset.

#### 6.5.2.3 3: Patient ages at presentation

The following is a dataset on skin sores in different communities in Australia and Oceania, specifically looking at the amount of time that passes between skin infections (Lydeamore et al. 2020a). This file includes multiple different datasets, and focuses on data from children in the first five years of their life, on househould visits, and on data collected during targeted studies (Lydeamore et al. 2020b).

```
url_file <- curl("https://doi.org/10.1371/journal.pcbi.1007838.s006")
ages <- url_file %>% read_csv()
```

```
Rows: 17150 Columns: 2
-- Column specification -------
Delimiter: ","
chr (1): dataset
dbl (1): time_difference
```

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

Let's see what this file is like real fast. We can use the command dim to see the rows and columns.

```
dim(ages)
```

```
[1] 17150 2
```

As you can see, this file has only two columns but 17,150 rows! For the column time\_difference, please calculate the mean, median, range, interquartile range, variance, standard deviation, coefficient of variation, standard error, and whether there are any "outliers".

**Synthesis question**: Folks will often think about probabilities of events being "low but never zero". What does that mean in the context of these data? What about these data make you feel like probabilities may decrease through time but never become zero?

## 7 Diagnosing data visually

### 7.1 The importance of visual inspection

Inspecting data visually can give us a lot of information about whether data are normally distributed and about whether there are any major errors or issues with our dataset. It can also help us determine if data meet model assumptions, or if we need to use different tests more appropriate for our datasets.

### 7.2 Sample data and preparation

First, we need to load our R libraries.

```
library(curl)
```

Using libcurl 8.7.1 with LibreSSL/3.3.6

-- Attaching core tidyverse packages ----

```
library(tidyverse)
```

```
v dplyr
 1.1.4
 v readr
 2.1.5
v forcats
 1.0.0
 v stringr
 1.5.1
v ggplot2 3.5.1
 v tibble
 3.2.1
v lubridate 1.9.3
 v tidyr
 1.3.1
v purrr
 1.0.2
-- Conflicts -----
 x dplyr::filter()
 masks stats::filter()
x dplyr::lag()
 masks stats::lag()
x readr::parse_date() masks curl::parse_date()
i Use the conflicted package (http://conflicted.r-lib.org/) to force all conflicts to become
```

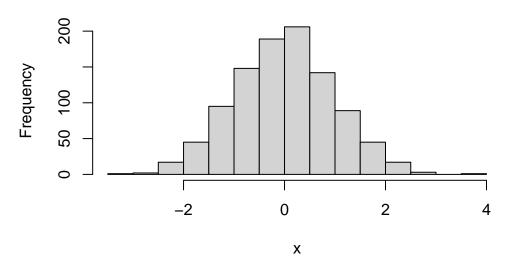
----- tidyverse 2.0.0 --

Next, we can download our data sample.

### 7.3 Histograms

A histogram is a frequency diagram that we can use to visually diagnose data and their distributions. We are going to examine a histogram using a random string of data. R can generate random (though, actually pseudorandom) strings of data on command, pulling them from different distributions. These distributions are pseudorandom because we can't actually program R to be random, so it starts from a wide variety of pseudorandom points.

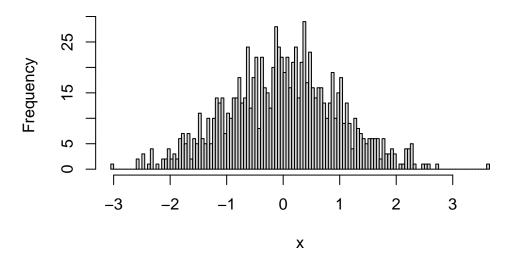
### Histogram of x



We can up the number of bins to see this better.

```
hist(x,breaks = 100)
```

## Histogram of x

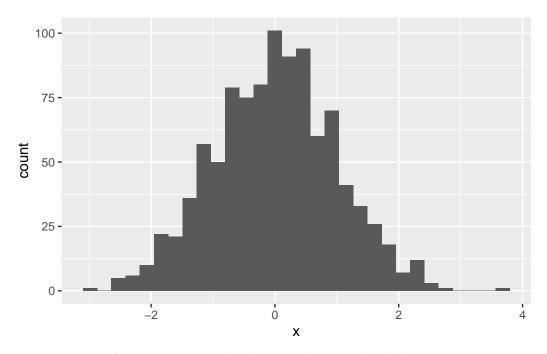


We can also use the program ggplot, part of the tidyverse, to create histograms.

```
ggplot requires data frames
x2 <- x %>% as.data.frame()
colnames(x2) <- "x"

ggplot(data = x2, aes(x = x)) +
 geom_histogram()</pre>
```

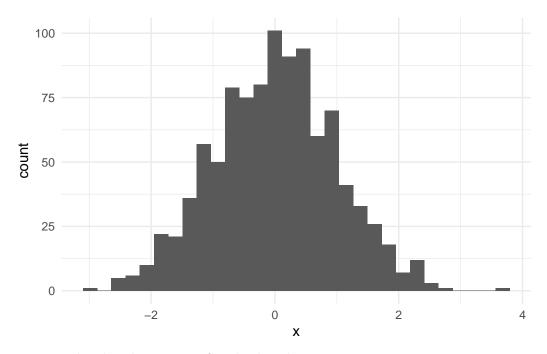
`stat\_bin()` using `bins = 30`. Pick better value with `binwidth`.



ggplot is nice because we can also clean up this graph a little.

```
ggplot(x2,aes(x=x)) + geom_histogram() +
 theme_minimal()
```

`stat\_bin()` using `bins = 30`. Pick better value with `binwidth`.

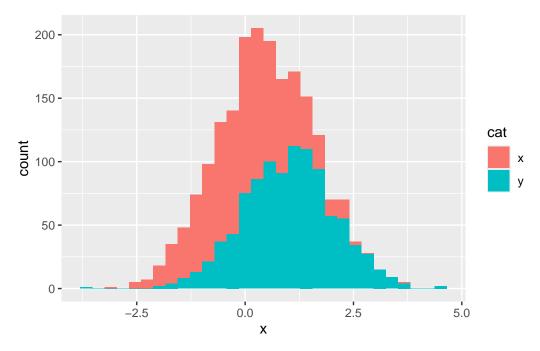


We can also do a histogram of multiple values at once in  ${\cal R}.$ 

```
x cat
1 -1.5737184 x
2 1.1922186 x
3 -0.5768628 x
4 -0.2449589 x
5 -1.2468854 x
6 -0.2774695 x
```

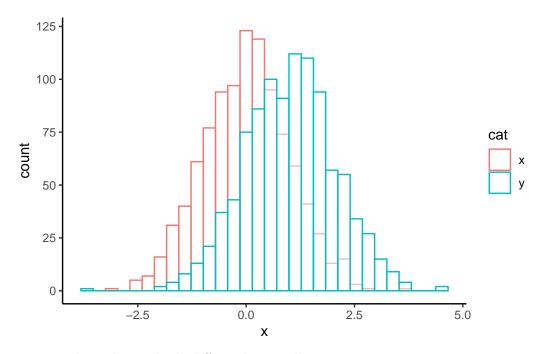
```
ggplot(xy,aes(x = x, fill = cat)) +
 geom_histogram()
```

`stat\_bin()` using `bins = 30`. Pick better value with `binwidth`.



We can also make this look a little nicer.

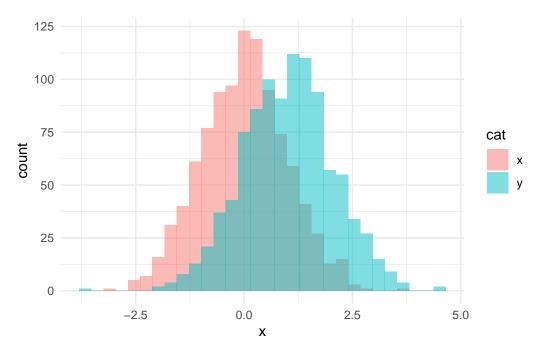
`stat\_bin()` using `bins = 30`. Pick better value with `binwidth`.



We can show these a little differently as well.

```
ggplot(xy, aes(x = x, fill = cat))+
 geom_histogram(position = "identity", alpha = 0.5) +
 theme_minimal()
```

`stat\_bin()` using `bins = 30`. Pick better value with `binwidth`.



There are lots of other commands you can incorporate as well if you so choose; I recommend checking sites like this one.

#### 7.4 Skewness

Skew is a measure of how much a dataset "leans" to the positive or negative directions (*i.e.*, to the "left" or to the "right"). To calculate skew, we are going to use the moments library.

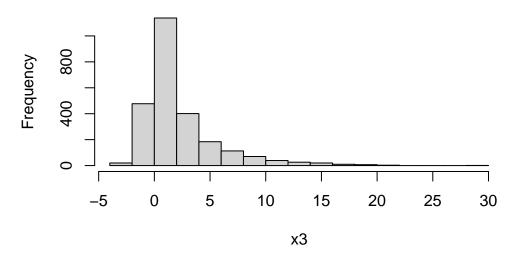
```
don't forget to install if needed!
library(moments)
skewness(x)
```

#### [1] 0.01887377

Generally, a value between -1 and +1 for skewness is "acceptable" and not considered overly skewed. Positive values indicate "right" skew and negative values indicate a "left" skew. If something is too skewed, it may violate assumptions of normality and thus need *non-parametric* tests rather than our standard parametric tests - something we will cover later!

Let's look at a skewed dataset. We are going to artificially create a skewed dataset from our  $\mathbf{x}$  vector.

### Histogram of x3



#### skewness(x3)

#### [1] 2.207081

As we can see, the above is a heavily skewed dataset with a positive ("right") skew.

### 7.5 Kurtosis

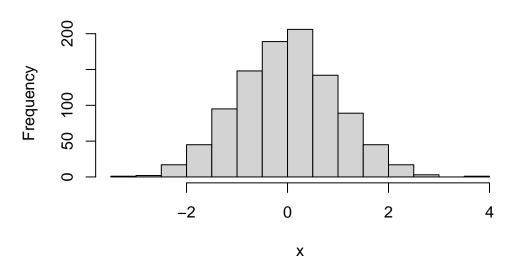
Kurtosis refers to how sharp or shallow the peak of the distribution is (platykurtic vs. leptokurtic). Remember - platykyrtic are plateaukurtic, wide and broad like a plateau, and leptokurtic distributions are sharp. Intermediate distributions that are roughly normal are mesokurtic.

Much like skewness, kurtosis values of > 2 and < -2 are generally considered extreme, and thus not mesokurtic. This threshold can vary a bit based on source, but for this class, we will use a threshold of  $\pm 2$  for both skewness and kurtosis.

Let's see the kurtosis of x. Note that when doing the equation, a normal distribution actually has a kurtosis of 3; thus, we are doing kurtosis -3 to "zero" the distribution and make it comparable to skewness.

#### hist(x)

### Histogram of x



# non-zeroed
kurtosis(x)

[1] 2.905307

# zeroed
kurtosis(x)-3

[1] -0.0946927

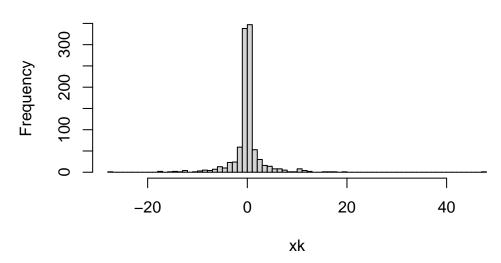
As expected, out values drawn from a normal distribution are not overly skewed. Let's compare these to a more kurtic distribution:

xk <- x<sup>3</sup>
kurtosis(xk)-3

[1] 42.9731

hist(xk,breaks = 100)

## Histogram of xk



As we can see, this is a very *leptokurtic* distribution.

## 7.6 Homework: Chapter 3

# 8 Normality & hypothesis testing

- 8.1 Normal distributions
- 8.2 Hypothesis testing
- 8.3 Homework: Chapter 8

# 9 Exam 2 practice

## 9.1 Exam 2

The following is practice for Exam 2.

## 10 Probability distributions

- 10.1 Probability distributions
- 10.2 Binomial distribution
- 10.3 Poisson distribution
- 10.4 Chi-square distribution
- 10.5 Fisher's exact test
- 10.6 Homework
- 10.6.1 Chapter 5
- 10.6.2 Chapter 7

# 11 Single population means testing

- 11.1 Introduction
- 11.2 *t*-distribution
- 11.3 *t*-tests
- 11.4 Wilcoxon tests
- 11.5 Confidence intervals
- 11.6 Homework: Chapter 9

# 12 Two sample tests

- 12.1 Introduction
- 12.2 *t*-tests
- 12.3 Mann-Whitney U tests
- 12.4 Error
- 12.5 Homework: Chapter 10

## 13 ANOVA: Part 1

- 13.1 Introduction
- 13.2 ANOVA: By hand
- 13.3 ANOVA: By *R*
- 13.4 Kruskal-Wallis tests
- 13.5 Homework: Chapter 11

## 14 ANOVA: Part 2

- 14.1 Two-way ANOVA
- 14.2 Designs
- 14.2.1 Randomized block design
- 14.2.2 Repeated measures
- 14.2.3 Factorial ANOVA
- 14.3 Friedman's test
- 14.4 Homework: Chapter 12

# 15 Correlation & regression

- 15.1 Introduction
- 15.2 Correlation
- 15.2.1 Pearson's
- 15.2.2 Spearman's
- 15.2.3 Other non-parametric methods
- 15.3 Correlation
- 15.3.1 Parametric
- 15.3.2 Non-parametric
- 15.4 Homework
- 15.4.1 Chapter 13
- 15.4.2 Chapter 14

# 16 Final exam & review

- 16.1 Pick the test
- 16.2 Final review

## 17 Conclusions

Parting thoughts about the course.

#### 17.1

(pronounced doh-dah-dah-go-huh-ee) is a traditional Cherokee farewell. It does not mean goodbye, but rather reflects a parting of ways until a group of folks meet again.

I enjoyed getting to know all of you in class, and please feel free to reach out or stop by and say hi if you are ever passing through Kearney in the future or if you need help with something biology related.

Wishing you the best,

Dr. Cooper

### References

- Cooper, J. C. (2021). Biogeographic and Ecologic Drivers of Avian Diversity. [Online.] Available at https://doi.org/10.6082/uchicago.3379.
- Lydeamore, M. J., P. T. Campbell, D. J. Price, Y. Wu, A. J. Marcato, W. Cuningham, J. R. Carapetis, R. M. Andrews, M. I. McDonald, J. McVernon, S. Y. C. Tong, and J. M. McCaw (2020b). Patient ages at presentation. https://doi.org/10.1371/journal.pcbi.1007838.s006
- Lydeamore, M. J., P. T. Campbell, D. J. Price, Y. Wu, A. J. Marcato, W. Cuningham, J. R. Carapetis, R. M. Andrews, M. I. McDonald, J. McVernon, S. Y. C. Tong, and J. M. McCaw (2020a). Estimation of the force of infection and infectious period of skin sores in remote Australian communities using interval-censored data. PLOS Computational Biology 16:e1007838.
- Moura, R., N. P. Santos, and A. Rocha (2023). Processed csv file of the piracy dataset. https://doi.org/10.6084/m9.figshare.24119643.v1