

My Final College Paper

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The Division of Biology
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I want to thank a few people.

Preface

This is an example of a thesis setup to use the reed thesis document class (for LaTeX) and the R bookdown package, in general.

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Abstract

The preface pretty much says it all.

Second paragraph of abstract starts here.

Dedication

You can have a dedication here if you wish.

Introduction

All dynamic biological processes require control over transcription, translation, or post-translational products. My work has focused on characterizing this control using RNA sequencing techniques, primarily on transcription and translation, to better understand how stem cell differentiation occurs. Stem cells are cells with the unique ability to develop into more specialized cell types in a process called differentiation. Some stem cell, including those focused on in my work, also have the ability to “self-renew,” a process that allows one stem cell to copy itself giving rise to two stem cells. These processes must be carefully balanced as excess self-renewal will result in cells that do not give rise to differentiated cells necessary for further development or biological function. However, excess differentiation will result in the lack of an available pool of stem cells, preventing future differentiation and development.

The decision of a stem cell to either self renew or differentiate is controlled by specific cellular pathways that can act at the level of transcription, translation, or post-translation. To study the regulation of these pathways in-vivo I have used the female *Drosophila* germline as a model system. The female *Drosophila* germline is contained within two pairs of ovaries. Ovaries consist of two main types of tissue, soma and germline. Each ovary is made up of strands called ovarioles. Ovarioles represent an assembly line of successive development. At the anterior tip of each ovariole a structure called a germarium is present. At the anterior of the germarium two to three stem cells are housed in a somatic niche. These germline stem cells (GSCs) can self-renew, or differentiate giving rise to a daughter cell called a cystoblast (CB). The CB turns on a differentiation factor called bag of marbles (bam). This CB then undergoes four incomplete cellular divisions, resulting in interconnected cysts consisting of two, four, eight, and finally sixteen cells. One of these cells is designated as the oocyte while the rest of the cells will become nurse cells. The sixteen cell cyst is then encapsulated by somatic cells, forming egg chambers. Egg chambers successively grow in size in fourteen stages. During this time the nurse cells produce mRNAs and proteins that are transported to the oocyte. The oocyte continues to grow, while the nurse cells eventually die, dumping their contents into the oocyte. Once the oocyte reaches the final, 14th stage it is known as an egg.

Each of the steps from GSC to egg require changes in cellular pathways. These changes can occur at the level of transcription, translation, or post-translation. Decades of research has elucidated many of the changes that occur during oogenesis, however, many players in this process still remain mysterious. My work has helped to identify and characterize novel developmental mechanisms that are required

for the successive developmental transitions that take place during oogenesis. I have leveraged RNAseq and polysome-seq to probe the global transcriptional and translational landscape over development. I have also used the power of *Drosophila* genetics in concert with these sequencing techniques to identify and characterize misregulated pathways.

Chapter 1

R Markdown Basics

Here is a brief introduction into using *R Markdown*. *Markdown* is a simple formatting syntax for authoring HTML, PDF, and MS Word documents. *R Markdown* provides the flexibility of *Markdown* with the implementation of **R** input and output. For more details on using *R Markdown* see <https://rmarkdown.rstudio.com>.

Be careful with your spacing in *Markdown* documents. While whitespace largely is ignored, it does at times give *Markdown* signals as to how to proceed. As a habit, try to keep everything left aligned whenever possible, especially as you type a new paragraph. In other words, there is no need to indent basic text in the Rmd document (in fact, it might cause your text to do funny things if you do).

1.1 Lists

It's easy to create a list. It can be unordered like

- Item 1
- Item 2

or it can be ordered like

1. Item 1
2. Item 2

Notice that I intentionally mislabeled Item 2 as number 4. *Markdown* automatically figures this out! You can put any numbers in the list and it will create the list. Check it out below.

To create a sublist, just indent the values a bit (at least four spaces or a tab). (Here's one case where indentation is key!)

1. Item 1
2. Item 2
3. Item 3
 - Item 3a
 - Item 3b

1.2 Line breaks

Make sure to add white space between lines if you'd like to start a new paragraph. Look at what happens below in the outputted document if you don't:

Here is the first sentence. Here is another sentence. Here is the last sentence to end the paragraph. This should be a new paragraph.

Now for the correct way:

Here is the first sentence. Here is another sentence. Here is the last sentence to end the paragraph.

This should be a new paragraph.

1.3 R chunks

When you click the **Knit** button above 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 (`cars` is a built-in **R** dataset):

```
summary(cars)
```

	speed		dist
Min.	: 4.0	Min.	: 2.00
1st Qu.	:12.0	1st Qu.	: 26.00
Median	:15.0	Median	: 36.00
Mean	:15.4	Mean	: 42.98
3rd Qu.	:19.0	3rd Qu.	: 56.00
Max.	:25.0	Max.	:120.00

1.4 Inline code

If you'd like to put the results of your analysis directly into your discussion, add inline code like this:

The `cos` of 2π is 1.

Another example would be the direct calculation of the standard deviation:

The standard deviation of `speed` in `cars` is 5.2876444.

One last neat feature is the use of the `ifelse` conditional statement which can be used to output text depending on the result of an **R** calculation:

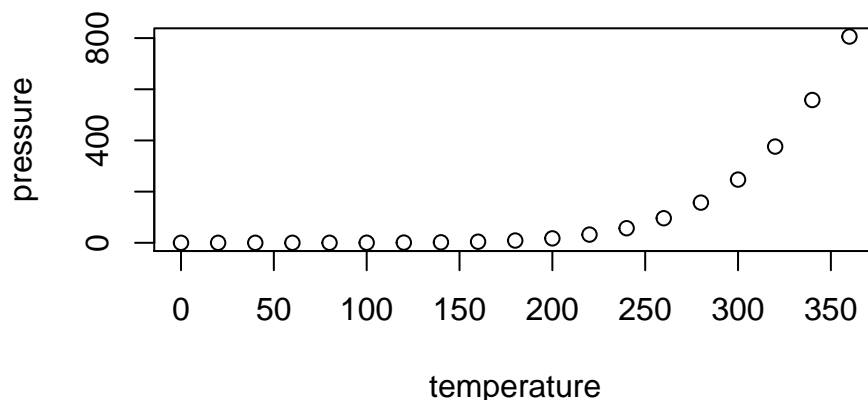
The standard deviation is less than 6.

Note the use of `>` here, which signifies a quotation environment that will be indented.

As you see with π above, mathematics can be added by surrounding the mathematical text with dollar signs. More examples of this are in Mathematics and Science if you uncomment the code in Math.

1.5 Including plots

You can also embed plots. For example, here is a way to use the base **R** graphics package to produce a plot using the built-in `pressure` dataset:



Note that the `echo=FALSE` parameter was added to the code chunk to prevent printing of the **R** code that generated the plot. There are plenty of other ways to add chunk options (like `fig.height` and `fig.width` in the chunk above). More information is available at <https://yihui.org/knitr/options/>.

Another useful chunk option is the setting of `cache=TRUE` as you see here. If document rendering becomes time consuming due to long computations or plots that are expensive to generate you can use knitr caching to improve performance. Later in this file, you'll see a way to reference plots created in **R** or external figures.

1.6 Loading and exploring data

Included in this template is a file called `flights.csv`. This file includes a subset of the larger dataset of information about all flights that departed from Seattle and Portland in 2014. More information about this dataset and its **R** package is available at <https://github.com/ismayc/pnwflights14>. This subset includes only Portland flights and only rows that were complete with no missing values. Merges were also done with the `airports` and `airlines` data sets in the `pnwflights14` package to get more descriptive airport and airline names.

We can load in this data set using the following commands:

```
# flights.csv is in the data directory
flights_path <- here::here("data", "flights.csv")
# string columns will be read in as strings and not factors now
flights <- read.csv(flights_path, stringsAsFactors = FALSE)
```

The data is now stored in the data frame called `flights` in **R**. To get a better

feel for the variables included in this dataset we can use a variety of functions. Here we can see the dimensions (rows by columns) and also the names of the columns.

```
dim(flights)
```

```
[1] 12649    16
```

```
names(flights)
```

```
[1] "month"      "day"        "dep_time"    "dep_delay"
[5] "arr_time"    "arr_delay"  "carrier"     "tailnum"
[9] "flight"     "dest"       "air_time"    "distance"
[13] "hour"       "minute"     "carrier_name" "dest_name"
```

Another good idea is to take a look at the dataset in table form. With this dataset having more than 20,000 rows, we won't explicitly show the results of the command here. I recommend you enter the command into the Console *after* you have run the **R** chunks above to load the data into **R**.

```
View(flights)
```

While not required, it is highly recommended you use the **dplyr** package to manipulate and summarize your data set as needed. It uses a syntax that is easy to understand using chaining operations. Below I've created a few examples of using **dplyr** to get information about the Portland flights in 2014. You will also see the use of the **ggplot2** package, which produces beautiful, high-quality academic visuals.

We begin by checking to ensure that needed packages are installed and then we load them into our current working environment:

```
# List of packages required for this analysis
pkg <- c("dplyr", "ggplot2", "knitr", "bookdown")
# Check if packages are not installed and assign the
# names of the packages not installed to the variable new.pkg
new.pkg <- pkg[!(pkg %in% installed.packages())]
# If there are any packages in the list that aren't installed,
# install them
if (length(new.pkg)) {
  install.packages(new.pkg, repos = "https://cran.rstudio.com")
}
# Load packages
library(thesisdown)
library(dplyr)
library(ggplot2)
library(knitr)
```


The example we show here does the following:

- Selects only the `carrier_name` and `arr_delay` from the `flights` dataset and then assigns this subset to a new variable called `flights2`.
- Using `flights2`, we determine the largest arrival delay for each of the carriers.

```
flights2 <- flights %>%
  select(carrier_name, arr_delay)
max_delays <- flights2 %>%
  group_by(carrier_name) %>%
  summarize(max_arr_delay = max(arr_delay, na.rm = TRUE))
```

A useful function in the `knitr` package for making nice tables in *R Markdown* is called `kable`. It is much easier to use than manually entering values into a table by copying and pasting values into Excel or LaTeX. This again goes to show how nice reproducible documents can be! (Note the use of `results="asis"`, which will produce the table instead of the code to create the table.) The `caption.short` argument is used to include a shorter title to appear in the List of Tables.

```
kable(max_delays,
  col.names = c("Airline", "Max Arrival Delay"),
  caption = "Maximum Delays by Airline",
  caption.short = "Max Delays by Airline",
  longtable = TRUE,
  booktabs = TRUE
)
```

Table 1.1: Maximum Delays by Airline

Airline	Max Arrival Delay
Alaska Airlines Inc.	338
American Airlines Inc.	1539
Delta Air Lines Inc.	371
Frontier Airlines Inc.	166
Hawaiian Airlines Inc.	116
JetBlue Airways	256
SkyWest Airlines Inc.	321
Southwest Airlines Co.	315
United Air Lines Inc.	319
US Airways Inc.	347
Virgin America	366

The last two options make the table a little easier-to-read.

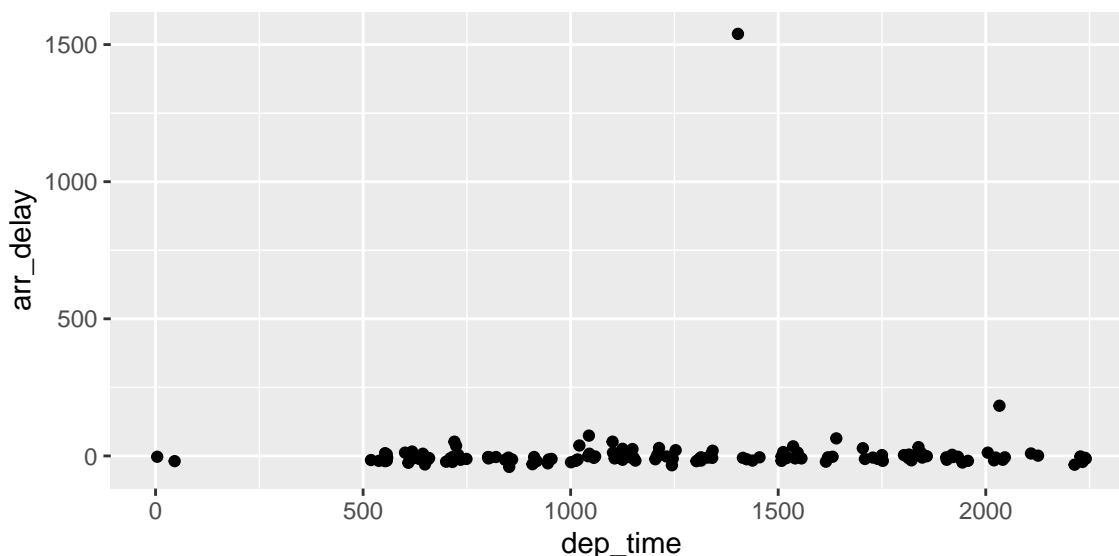
We can further look into the properties of the largest value here for American Airlines Inc. To do so, we can isolate the row corresponding to the arrival delay of 1539 minutes for American in our original `flights` dataset.

```
flights %>%
  filter(
    arr_delay == 1539,
    carrier_name == "American Airlines Inc."
  ) %>%
  select(-c(
    month, day, carrier, dest_name, hour,
    minute, carrier_name, arr_delay
  ))
```

```
dep_time dep_delay arr_time tailnum flight dest air_time distance
1      1403      1553      1934  N595AA   1568  DFW        182      1616
```

We see that the flight occurred on March 3rd and departed a little after 2 PM on its way to Dallas/Fort Worth. Lastly, we show how we can visualize the arrival delay of all departing flights from Portland on March 3rd against time of departure.

```
flights %>%
  filter(month == 3, day == 3) %>%
  ggplot(aes(x = dep_time, y = arr_delay)) +
  geom_point()
```



1.7 Additional resources

- *Markdown* Cheatsheet - <https://github.com/adam-p/markdown-here/wiki/Markdown-Cheatsheet>
- *R Markdown*
 - Reference Guide - <https://www.rstudio.com/wp-content/uploads/2015/03/rmarkdown-reference.pdf>
 - Cheatsheet - <https://github.com/rstudio/cheatsheets/raw/master/rmarkdown-2.0.pdf>
- *RStudio IDE*
 - Cheatsheet - <https://github.com/rstudio/cheatsheets/raw/master/rstudio-ide.pdf>
 - Official website - <https://rstudio.com/products/rstudio/>
- Introduction to dplyr - <https://cran.rstudio.com/web/packages/dplyr/vignettes/dplyr.html>
- ggplot2
 - Documentation - <https://ggplot2.tidyverse.org/>
 - Cheatsheet - <https://github.com/rstudio/cheatsheets/raw/master/data-visualization-2.1.pdf>

Chapter 2

Mathematics and Science

2.1 Math

T_EX is the best way to typeset mathematics. Donald Knuth designed T_EX when he got frustrated at how long it was taking the typesetters to finish his book, which contained a lot of mathematics. One nice feature of *R Markdown* is its ability to read LaTeX code directly.

If you are doing a thesis that will involve lots of math, you will want to read the following section which has been commented out. If you're not going to use math, skip over or delete this next commented section.

2.2 Chemistry 101: Symbols

Chemical formulas will look best if they are not italicized. Get around math mode's automatic italicizing in LaTeX by using the argument `$\mathrm{formula here}$` , with your formula inside the curly brackets. (Notice the use of the backticks here which enclose text that acts as code.)

So, Fe₂²⁺Cr₂O₄ is written `$\mathrm{Fe_2^{2+}Cr_2O_4}$` .

Exponent or Superscript: O⁻

Subscript: CH₄

To stack numbers or letters as in Fe₂²⁺, the subscript is defined first, and then the superscript is defined.

Bullet: CuCl • 7H₂O

Delta: Δ

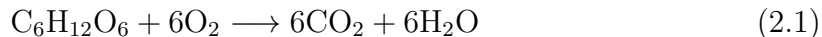
Reaction Arrows: \longrightarrow or $\xrightarrow{\text{solution}}$

Resonance Arrows: \longleftrightarrow

Reversible Reaction Arrows: \rightleftharpoons

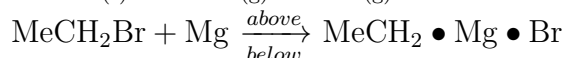
2.2.1 Typesetting reactions

You may wish to put your reaction in an equation environment, which means that LaTeX will place the reaction where it fits and will number the equations for you.



We can reference this combustion of glucose reaction via Equation (2.1).

2.2.2 Other examples of reactions



2.3 Physics

Many of the symbols you will need can be found on the math page <https://web.reed.edu/cis/help/latex/math.html> and the Comprehensive LaTeX Symbol Guide (<https://mirror.utexas.edu/ctan/info/symbols/comprehensive/symbols-letter.pdf>).

2.4 Biology

You will probably find the resources at <https://www.lecb.ncifcrf.gov/~toms/latex.html> helpful, particularly the links to bst files for various journals. You may also be interested in TeXShade for nucleotide typesetting (<https://homepages.uni-tuebingen.de/beitz/txe.html>). Be sure to read the proceeding chapter on graphics and tables.

Chapter 3

Graphics, References, and Labels

3.1 Figures

If your thesis has a lot of figures, *R Markdown* might behave better for you than that other word processor. One perk is that it will automatically number the figures accordingly in each chapter. You'll also be able to create a label for each figure, add a caption, and then reference the figure in a way similar to what we saw with tables earlier. If you label your figures, you can move the figures around and *R Markdown* will automatically adjust the numbering for you. No need for you to remember! So that you don't have to get too far into LaTeX to do this, a couple **R** functions have been created for you to assist. You'll see their use below.

In the **R** chunk below, we will load in a picture stored as `reed.jpg` in our main directory. We then give it the caption of "Reed logo," the label of "reedlogo," and specify that this is a figure. Make note of the different **R** chunk options that are given in the R Markdown file (not shown in the knitted document).

```
include_graphics(path = "figure/reed.jpg")
```



Figure 3.1: Reed logo

Here is a reference to the Reed logo: Figure 3.1. Note the use of the `fig:` code here. By naming the **R** chunk that contains the figure, we can then reference that figure later as done in the first sentence here. We can also specify the caption for the figure via the R chunk option `fig.cap`.

Below we will investigate how to save the output of an **R** plot and label it in a way similar to that done above. Recall the `flights` dataset from Chapter 1. (Note that we've shown a different way to reference a section or chapter here.) We will next explore a bar graph with the mean flight departure delays by airline from Portland for 2014.

```
mean_delay_by_carrier <- flights %>%  
  group_by(carrier) %>%  
  summarize(mean_dep_delay = mean(dep_delay))  
ggplot(mean_delay_by_carrier, aes(x = carrier, y = mean_dep_delay)) +  
  geom_bar(position = "identity", stat = "identity", fill = "red")
```

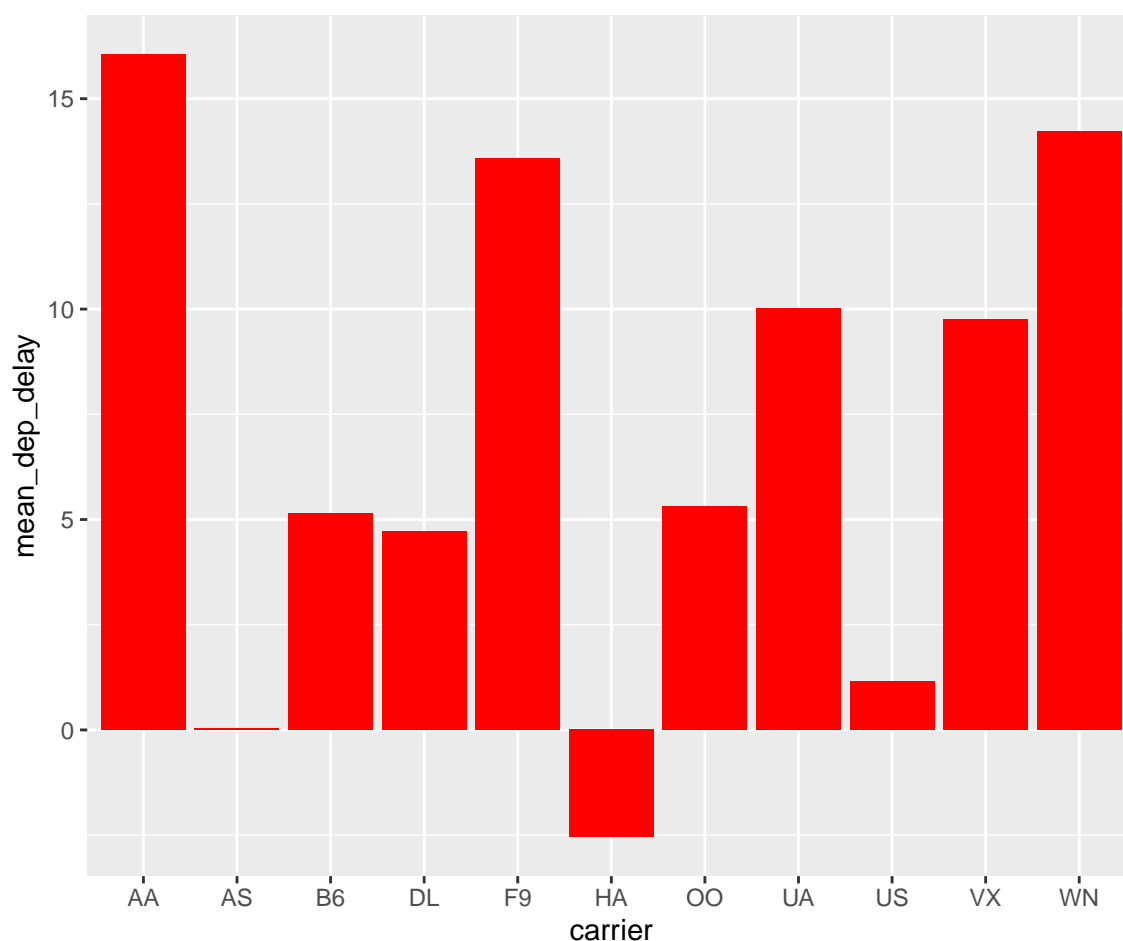


Figure 3.2: Mean Delays by Airline

Here is a reference to this image: Figure 3.2.

A table linking these carrier codes to airline names is available at <https://github.com/ismayc/pnwflights14/blob/master/data/airlines.csv>.

Next, we will explore the use of the `out.extra` chunk option, which can be used to shrink or expand an image loaded from a file by specifying `"scale= "`. Here we use the mathematical graph stored in the “subdivision.pdf” file.

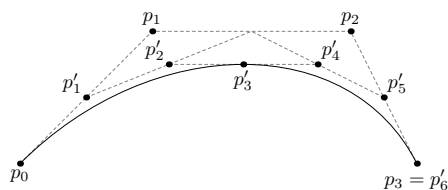


Figure 3.3: Subdiv. graph

Here is a reference to this image: Figure 3.3. Note that `echo=FALSE` is specified so that the **R** code is hidden in the document.

More Figure Stuff

Lastly, we will explore how to rotate and enlarge figures using the `out.extra` chunk option. (Currently this only works in the PDF version of the book.)

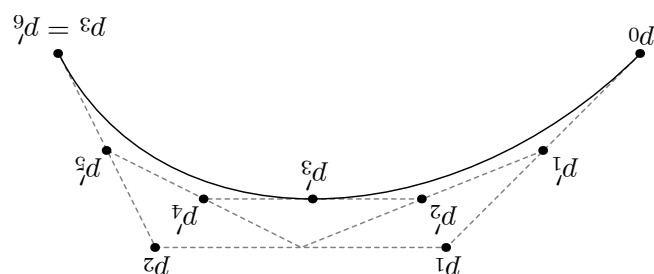


Figure 3.4: A Larger Figure, Flipped Upside Down

As another example, here is a reference: Figure 3.4.

3.2 Footnotes and Endnotes

You might want to footnote something.¹ The footnote will be in a smaller font and placed appropriately. Endnotes work in much the same way. More information can be found about both on the CUS site or feel free to reach out to `data@reed.edu`.

3.3 Bibliographies

Of course you will need to cite things, and you will probably accumulate an armful of sources. There are a variety of tools available for creating a bibliography database (stored with the `.bib` extension). In addition to BibTeX suggested below, you may want to consider using the free and easy-to-use tool called Zotero. The Reed librarians have created Zotero documentation at <https://libguides.reed>.

¹footnote text

edu/citation/zotero. In addition, a tutorial is available from Middlebury College at <https://sites.middlebury.edu/zoteromiddlebury/>.

R Markdown uses *pandoc* (<https://pandoc.org/>) to build its bibliographies. One nice caveat of this is that you won't have to do a second compile to load in references as standard LaTeX requires. To cite references in your thesis (after creating your bibliography database), place the reference name inside square brackets and precede it by the “at” symbol. For example, here's a reference to a book about worrying: (Molina & Borkovec, 1994). This `Molina1994` entry appears in a file called `thesis.bib` in the `bib` folder. This bibliography database file was created by a program called BibTeX. You can call this file something else if you like (look at the YAML header in the main `.Rmd` file) and, by default, is to placed in the `bib` folder.

For more information about BibTeX and bibliographies, see our CUS site (<https://web.reed.edu/cis/help/latex/index.html>)². There are three pages on this topic: *bibtex* (which talks about using BibTeX, at <https://web.reed.edu/cis/help/latex/bibtex.html>), *bibtexstyles* (about how to find and use the bibliography style that best suits your needs, at <https://web.reed.edu/cis/help/latex/bibtexstyles.html>) and *bibman* (which covers how to make and maintain a bibliography by hand, without BibTeX, at <https://web.reed.edu/cis/help/latex/bibman.html>). The last page will not be useful unless you have only a few sources.

If you look at the YAML header at the top of the main `.Rmd` file you can see that we can specify the style of the bibliography by referencing the appropriate csl file. You can download a variety of different style files at <https://www.zotero.org/styles>. Make sure to download the file into the `csl` folder.

Tips for Bibliographies

- Like with thesis formatting, the sooner you start compiling your bibliography for something as large as thesis, the better. Typing in source after source is mind-numbing enough; do you really want to do it for hours on end in late April? Think of it as procrastination.
- The cite key (a citation's label) needs to be unique from the other entries.
- When you have more than one author or editor, you need to separate each author's name by the word “and” e.g. `Author = {Noble, Sam and Youngberg, Jessica},.`
- Bibliographies made using BibTeX (whether manually or using a manager) accept LaTeX markup, so you can italicize and add symbols as necessary.
- To force capitalization in an article title or where all lowercase is generally used, bracket the capital letter in curly braces.
- You can add a Reed Thesis citation³ option. The best way to do this is to use the `phdthesis` type of citation, and use the optional “type” field to enter “Reed thesis” or “Undergraduate thesis.”

²Reed College (2007)

³Noble (2002)

3.4 Anything else?

If you'd like to see examples of other things in this template, please contact the Data @ Reed team (email data@reed.edu) with your suggestions. We love to see people using *R Markdown* for their theses, and are happy to help.

Conclusion

If we don't want Conclusion to have a chapter number next to it, we can add the `{-}` attribute.

More info

And here's some other random info: the first paragraph after a chapter title or section head *shouldn't be* indented, because indents are to tell the reader that you're starting a new paragraph. Since that's obvious after a chapter or section title, proper typesetting doesn't add an indent there.

Appendix A

The First Appendix

This first appendix includes all of the R chunks of code that were hidden throughout the document (using the `include = FALSE` chunk tag) to help with readability and/or setup.

In the main Rmd file

```
# This chunk ensures that the thesisdown package is  
# installed and loaded. This thesisdown package includes  
# the template files for the thesis.  
if (!require(remotes)) {  
  if (params$`Install needed packages for {thesisdown}`) {  
    install.packages("remotes", repos = "https://cran.rstudio.com")  
  } else {  
    stop(  
      paste('You need to run install.packages("remotes")',  
            "first in the Console.")  
    )  
  }  
}  
  
if (!require(thesisdown)) {  
  if (params$`Install needed packages for {thesisdown}`) {  
    remotes::install_github("ismayc/thesisdown")  
  } else {  
    stop(  
      paste(  
        "You need to run",  
        'remotes::install_github("ismayc/thesisdown")',  
        "first in the Console."  
      )  
    )  
  }  
}  
  
library(thesisdown)
```

```
# Set how wide the R output will go  
options(width = 70)
```

In Chapter 3:

```
# This chunk ensures that the thesisdown package is  
# installed and loaded. This thesisdown package includes  
# the template files for the thesis and also two functions  
# used for labeling and referencing  
if (!require(remotes)) {  
  if (params$`Install needed packages for {thesisdown}`) {  
    install.packages("remotes", repos = "https://cran.rstudio.com")  
  } else {  
    stop(  
      paste(  
        'You need to run install.packages("remotes")',  
        "first in the Console."  
      )  
    )  
  }  
}  
if (!require(dplyr)) {  
  if (params$`Install needed packages for {thesisdown}`) {  
    install.packages("dplyr", repos = "https://cran.rstudio.com")  
  } else {  
    stop(  
      paste(  
        'You need to run install.packages("dplyr")',  
        "first in the Console."  
      )  
    )  
  }  
}  
if (!require(ggplot2)) {  
  if (params$`Install needed packages for {thesisdown}`) {  
    install.packages("ggplot2", repos = "https://cran.rstudio.com")  
  } else {  
    stop(  
      paste(  
        'You need to run install.packages("ggplot2")',  
        "first in the Console."  
      )  
    )  
  }  
}
```



```
if (!require(bookdown)) {  
  if (params$`Install needed packages for {thesisdown}`) {  
    install.packages("bookdown", repos = "https://cran.rstudio.com")  
  } else {  
    stop(  
      paste(  
        'You need to run install.packages("bookdown")',  
        "first in the Console."  
      )  
    )  
  }  
}  
  
if (!require(thesisdown)) {  
  if (params$`Install needed packages for {thesisdown}`) {  
    remotes::install_github("ismayc/thesisdown")  
  } else {  
    stop(  
      paste(  
        "You need to run",  
        'remotes::install_github("ismayc/thesisdown")',  
        "first in the Console."  
      )  
    )  
  }  
}  
  
library(thesisdown)  
library(dplyr)  
library(ggplot2)  
library(knitr)  
flights <- read.csv("data/flights.csv", stringsAsFactors = FALSE)
```


Appendix B

The Second Appendix, for Fun

References

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