# Reproducible data analysis using R and RStudio

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Data Science for Developing Scholars in Down Syndrome Research (DS3) 2025







#### Useful links for this session

#### https://github.com/DS3-2025/installing\_updating\_R-RStudio

https://docs.posit.co/ide/user/ide/get-started/
https://rstudio.github.io/cheatsheets/base-r.pdf
https://posit.co/download/rstudio-desktop/
https://rstudio.github.io/cheatsheets/html/rstudio-ide.html

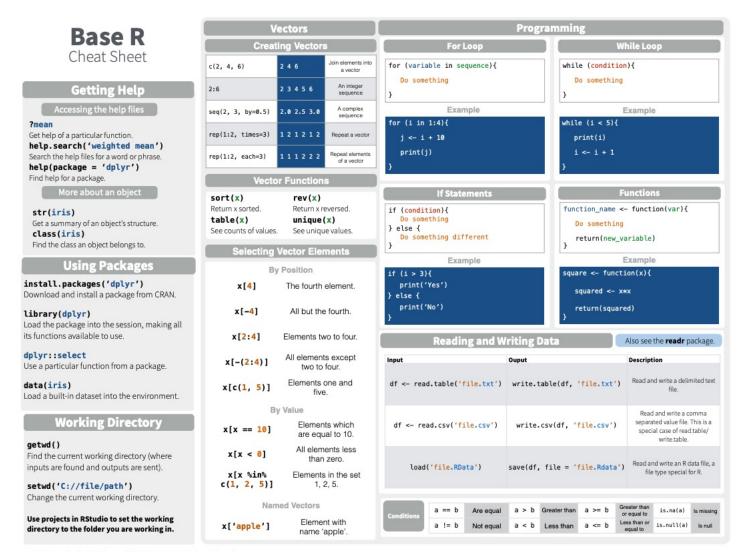
#### https://github.com/DS3-2025/Rproject\_template

https://support.rstudio.com/hc/en-us/articles/200526207-Using-RStudio-Projects
https://r4ds.had.co.nz/workflow-projects.html

https://tidyverse.tidyverse.org/

https://ggplot2.tidyverse.org/
https://ggplot2-book.org/





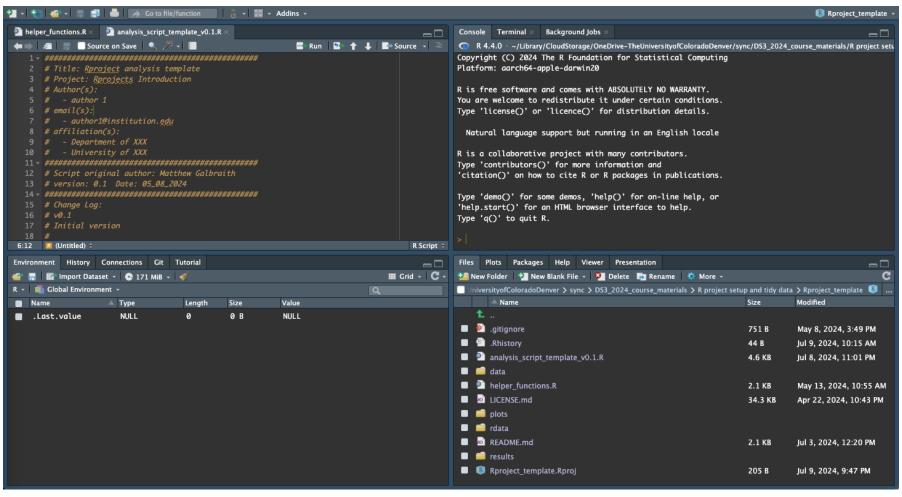
RStudio\* is a trademark of RStudio, Inc. • CC BY Mhairi McNeill • mhairihmcneill@gmail.com

Learn more at web page or vignette + package version + Updated: 3/15



#### Integrated Development Environment (IDE) for R

- Edit + execute R code
- Syntax highlighting, code completion, debugging
- View output, plots, help, environment

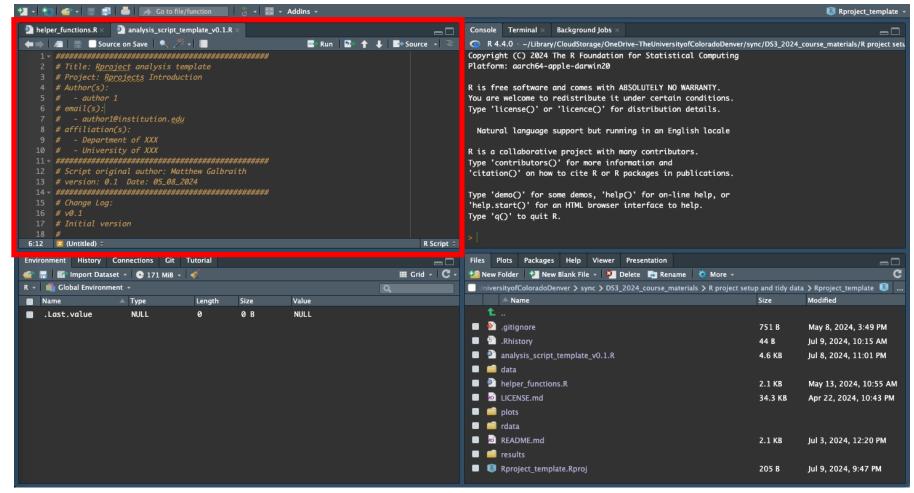




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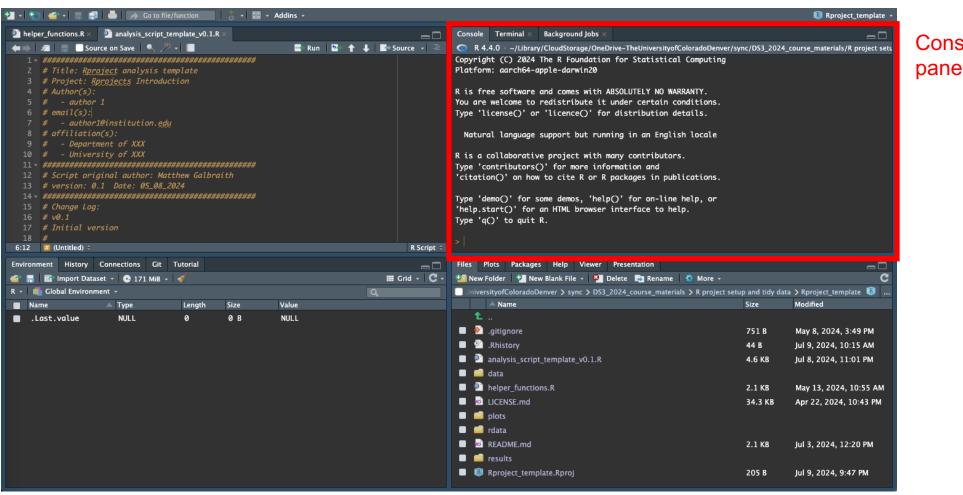
Source pane





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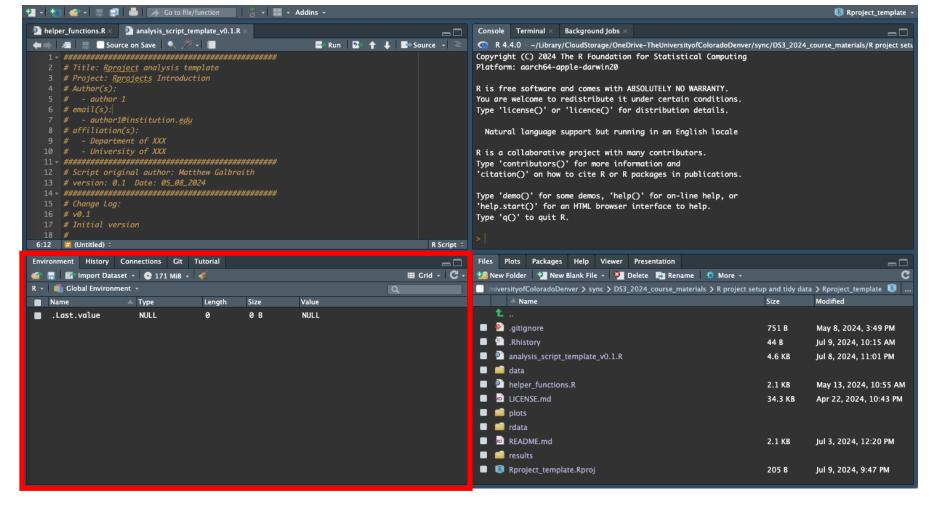


Console



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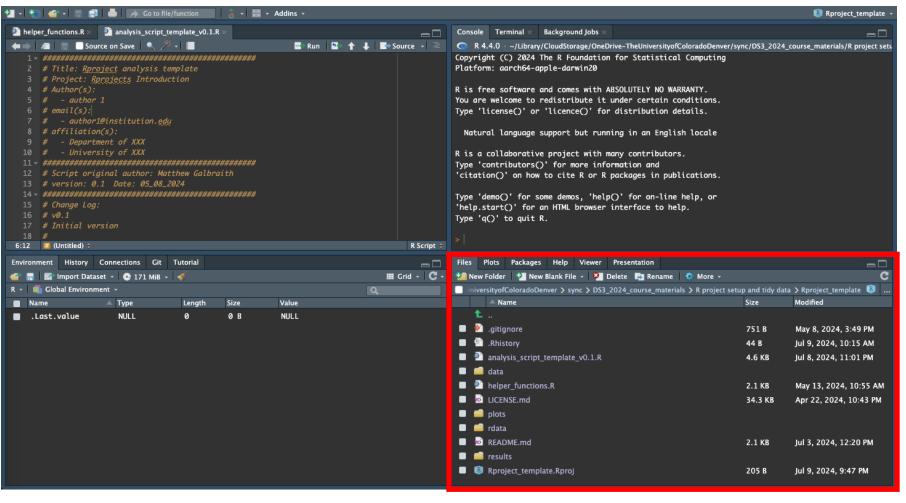


Environment etc pane



#### Integrated Development Environment (IDE) for R

- Edit + execute R code
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Files, Plots, Help etc pane

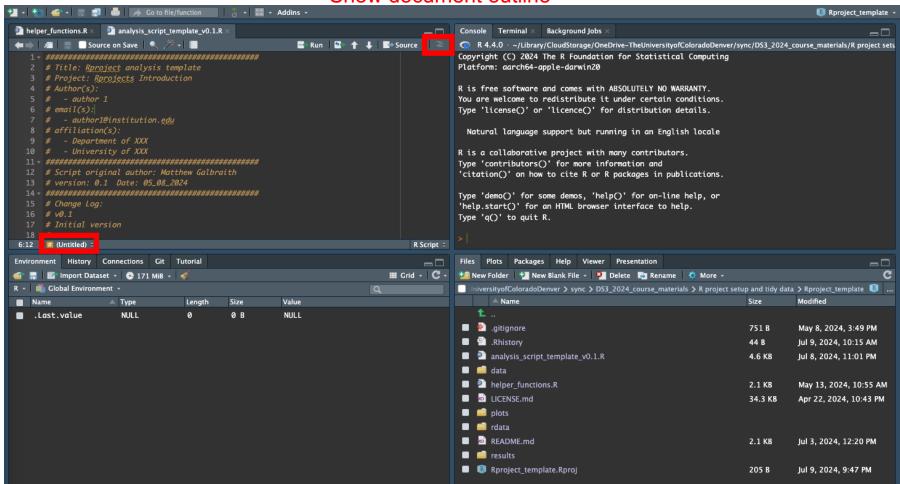
#### Rstudio – Code Sections

#### Staying organized with longer R scripts

Markdown-style comment headers, with the label followed by four or more dashes

```
# Section ----
## Subsection ----
### Sub-subsection ----
```

Show document outline



`Jump To` menu

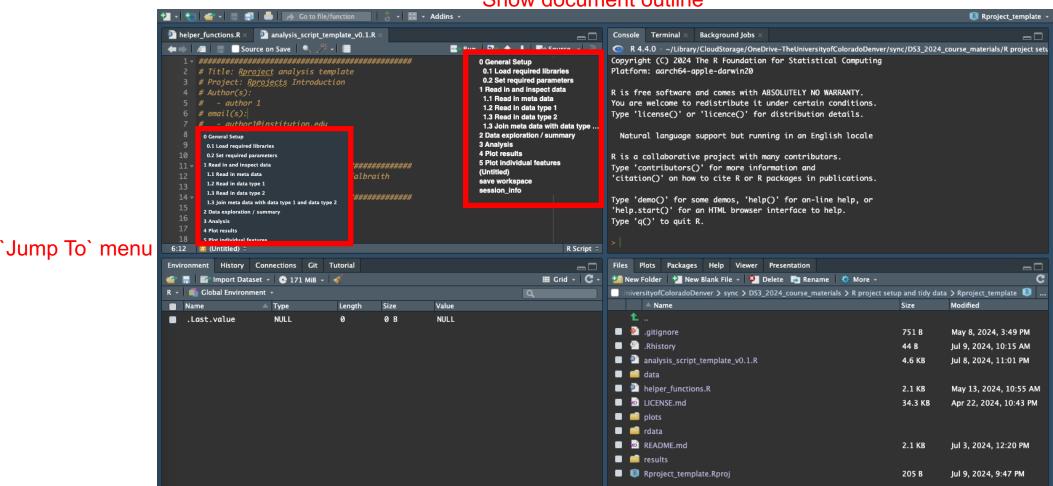
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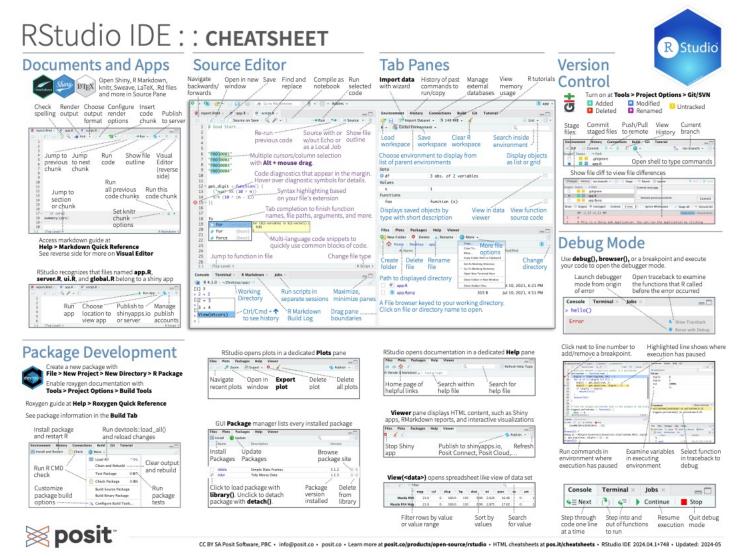
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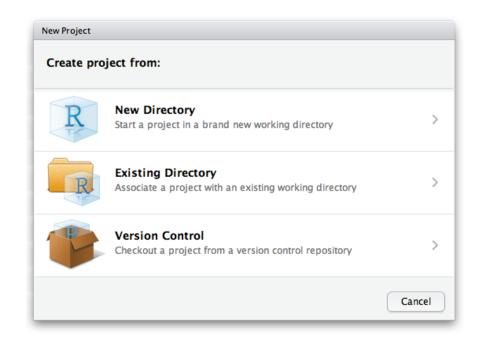
https://support.posit.co/hc/en-us/articles/200484568-Code-Folding-and-Sections-in-the-RStudio-IDE

#### **RStudio cheatsheet**



https://rstudio.github.io/cheatsheets/html/rstudio-ide.html

# Reproducible data analysis: RStudio Projects



#### **Project\_directory**

- -/data
- /results
- -/plots
- /rdata
- analysis\_script.R
- helper\_functions.R
- project.Rproj

- Usually open project via .Rproj file
- Automatically sets your working directory
- Self-contained set of directories, scripts, and data files (very important for multiple projects)

#### **Organizing your Rstudio Projects**

- Only /data and R scripts are required everything else can be recreated (incl. earlier versions)
- Treat /data directory as read-only
- Analysis outputs go to /results or /plots (with version info)
- R workspace and large intermediate files stored in /rdata
- Additional directories added as needed, eg /Archive
- Compatible with manual or other version control

# Reproducible data analysis: Package management using renv



- install.packages("renv")
- renv::init() to initialize a new project-local environment with a private R library
- renv::install() to install packages after initialization
- renv::snapshot() to save the state of your project to renv.lock
- renv::restore() to restore the state of your project from renv.lock

# Project\_directory - /data - /results - /plots - /rdata - /renv - renv.lock

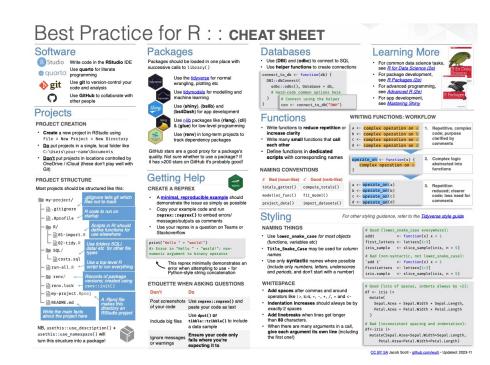
helper functions.R

analysis script.R

Allows for fully self-contained R projects (Usually) takes care of installing packages

# Reproducible data analysis: Best Practice

- Main analysis R script with standardized workflow (see next slides)
- Write functions to reduce repetition or increase clarity
- Define functions in dedicated script(s) (e.g. helper\_functions.R)
   keeps main script workflow clean + makes editing easier
- Comment your code!! (but don't just write what the code is doing)
- Adopt a coding style for consistency (e.g. <u>Tidyverse style guide</u>)
- Track versions and log changes + filenames with versions
- Inspect the data!!!!
- Check each step!!
- Visualize your data!!!!
- Keep track of any random seeds used
- Optional: use R notebooks/R markdown/Quarto/Roxygen to allow generation of reports with code (html, pdf, etc)



#### Title, Project, Author(s)

```
# Title: Rproject analysis template
# Project: Rprojects Introduction
 Author(s):
  - author 1
# email(s):
  author1@institution.edu
# affiliation(s):

    Department of XXX

  - University of XXX
# Script original author: Matthew Galbraith
# version: 0.1 Date: 05_08_2024
# Change Log:
# v0.1
# Initial version
```

#### Keep a change log

- What was changed and why?
- When?

Explain the purpose of the script + data types used

```
### Summary:
# Description of data wrangling and/or analysis being performed.
### Data type(s):
    A. Meta data
       Where/who did this data come from?
       What is the source of the original data and where is it stored?
    B. Data type 1
       Where/who did this data come from?
       What is the source of the original data and where is it stored?
    C. Data type 2
       Where/who did this data come from?
       What is the source of the original data and where is it stored?
### Workflow:
   1. Step 1 description
   2. Step 2 description
   3. Step 3 description
    4. Step 4 description
## Comments:
   Any further relevant details?
```

Outline the workflow steps (especially for longer scripts)

Load package libraries and custom functions

Packages should be loaded in one place with successive calls to library()

```
# 0 General Setup ----
# Initialize and install packages with renv
# renv::init()
## 0.1 Load required libraries ----
library("tidyverse")
library("readxl") # read .xlsx files
library("openxlsx") # data export to Excel workbooks
library("skimr") # data summary and validation
library("janitor") # data cleaning etc
library("ggrepel") # labelling points in plots
library("ggforce") # sina plots etc
library("patchwork") # arranging plots
library("tidyHeatmap") # tidy interface to ComplexHeatmap
library("plotly") # generating interactive plots
library("tictoc") # timer
library("conflicted") # force all conflicts to become errors
conflict_prefer("filter", "dplyr")
conflict_prefer("select", "dplyr")
conflict_prefer("count", "dplyr")
library("here") # generate path to current project directory
source(here("helper_functions.R")) # load helper functions
```

Use comments to explain what packages are being used for Note use of the *here* package which defines and stores project directory path Note use of the *conflicted* package which helps manage function conflicts

Define file locations and other global variables (all input files should be in /data)

```
## 0.2 Set required parameters ----
# Input data files
meta_data_file <- here("data", "meta_data_file.txt") # comments/notes on this file?
data_type1_file <- here("data", "data_type1_file.txt") # comments/notes on this file?
data_type2__file <- here("data", "data_type1_file.txt") # comments/notes on this file?
# Other parameters
standard_colors <- c("Group1" = "#F8766D", "Group2" = "#00BFC4")
out_file_prefix <- "analysis_script_template_v0.1_"
# End required parameters ###</pre>
```

All plots and results exports should use *out\_file\_prefix* (and go to /plots or /results) Include script version number in *out\_file\_prefix* 

Read in and inspect meta data (sample and/or experiment information)

```
# 1 Read in and inspect data ----
## 1.1 Read in meta data ----
meta_data <- meta_data_file |>
    read_tsv() |>
    janitor::clean_names(case = "none")
# inspect
meta_data
meta_data |> skimr::skim()
#
```

How many samples do you expect? How many measurements do you expect? Which identifiers are unique?

#### Read in and inspect your main data

```
## 1.2 Read in data type 1 ----
data_type1 <- data_type1_file |>
    read_tsv() |>
    janitor::clean_names(case = "none")
# inspect
data_type1
data_type1 |> skimr::skim()
#

## 1.3 Read in data type 2 ----
data_type2 <- data_type2_file |>
    read_tsv() |>
    janitor::clean_names(case = "none")
# inspect
data_type2
data_type2 |> skimr::skim()
#
```

How many samples do you expect? How many measurements do you expect? Which identifiers are unique?

Join meta data with main data

```
## 1.3 Join meta data with data type 1 and data type 2 ----
combined <- meta_data |>
  inner_join(data_type1) |>
  inner_join(data_type2)
# check number of rows returned
```

Do you have all sample rows after join? CHECK

#### Analysis specific sections:

- Data exploration
- Statistical testing
- Plot results
- Export results
- Plot individual features

```
# 2 Data exploration / summary ----
# check data distribution(s), outliers etc

# 3 Analysis ----
# statistical testing
# assemble and export results

# 4 Plot results ----
# plot results summaries

# 5 Plot individual features ----
# plot interesting/significant features
```

Finally: save and/or reload workspace

This can save time when resuming, but make sure to resave if something is changed or errors fixed!!

# Into the Tidyverse



https://tidyverse.tidyverse.org/

Base R: install.packages("tidyverse")

renv: renv::install("tidyverse")

#### Core tidyverse packages:

ggplot2, for data visualisation.

dplyr, for data manipulation.

tidyr, for data tidying.

readr, for data import.

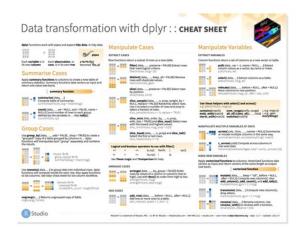
purrr, for functional programming.

tibble, for tibbles, a modern re-imagining of data frames.

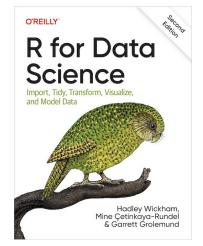
stringr, for strings.

forcats, for factors.

lubridate, for date/times.







# Into the Tidyverse: Pipes



#### **Pipe operator** %>%

- Avoids nesting
- Minimizes need to create objects and functions
- Structure sequences of operations left-to-right or top-to-bottom
- Easy to inspect and add steps anywhere
- x %>% f is equivalent to f(x)
- x %>% f(y) is equivalent to f(x, y)
- x %>% f %>% g %>% h is equivalent to h (g(f(x)))
- Keyboard shortcut in RStudio: cmd/ctrl+shift+m
- Note: R 4.1.0 introduced a native pipe operator |>
   with some minor differences
   https://www.tidyverse.org/blog/2023/04/base-vs-magrittr-pipe/

# Into the Tidyverse: syntax

#### Base R "dollar sign" syntax

Example - summary statistics:

#### one continuous variable:

mean (mtcars\$mpg)

#### one categorical variable:

table (mtcars\$cyl)

#### two categorical variables:

table(mtcars\$cyl, mtcars\$am)

#### one continuous, one categorical:

```
mean (mtcars$mpg[mtcars$cyl==4])
mean (mtcars$mpg[mtcars$cyl==6])
mean (mtcars$mpg[mtcars$cyl==8])
```

#### Tidyverse syntax

Example - summary statistics:

#### one continuous variable:

```
mtcars %>% dplyr::summarize(mean(mpg))
one categorical variable:
```

```
mtcars %>%
    dplyr::group_by(cyl)%>%
    dplyr::summarize(n())
```

#### two categorical variables:

```
mtcars %>%
    dplyr::group_by(cyl, am) %>%
    dplyr::summarize(n())
```

#### one continuous, one categorical:

```
mtcars %>%
    dplyr::group_by(cyl)%>%
    dplyr::summarize(mean(mpg))
```

# Into the Tidyverse

#### **Z-score calculation with base R:**

```
x <- sweep(sweep(t(dat), 1,
apply(t(dat),1, mean, na.rm=T), FUN = "-"),
1, apply(t(dat),1,sd,na.rm=T), FUN = "/")
```

- hard to decipher (learning barrier)
- have to enter target object name in several places

#### **Z-score calc with tidyverse + scale():**

```
zscores <- dat |>
      select(LabID, Analyte, Value) |>
      pivot wider() |>
      scale()
```

 Somewhat easier to decipher, but not obvious that this calculates Z-scores, even looking at ?scale defaults (center = TRUE, scale = TRUE)

#### **Manual Z-score calc with tidyverse:**

```
zscores <- dat |>
      select(LabID, Analyte, Value) |>
      group by(Analyte) |>
      mutate(
       zscore = (Value - mean(Value, na.rm = TRUE)) / sd(Value, na.rm = TRUE)
      ) |>
      ungroup()
```

- Naming of new variable
- Easier to see how calculation was performed
- Easy to keep both original and transformed values for comparison

# Into the Tidyverse

#### **Z-score calculation with base R:**

```
x <- sweep(sweep(t(dat), 1,
apply(t(dat),1,mean,na.rm=T), FUN = "-"),
1, apply(t(dat),1,sd,na.rm=T), FUN = "/")</pre>
```

#### **Even more verbose tidyverse version:**

```
zscores <- dat |>
    select(LabID, Analyte, Value) |>
    group_by(Analyte) |>
    mutate(
        mean = mean(Value, na.rm = TRUE),
        sd = sd(Value, na.rm = TRUE),
        zscore = (Value - mean) / sd
    ) |>
        ungroup()
```

- hard to decipher (learning barrier)
- have to enter target object name in several places

- Naming of new variable
- Easier to see how calculation was performed
- Easy to keep both original and transformed values for comparison







#### **Tibbles = enhanced data frames**

- Easier preview of data
- Concise summary information including data types

#### Importing delimited data

- Easy reading in of data from .txt, .csv, .tsv, .xlsx
- Guessing of column types
- Will not convert strings
- Imported as tibble

_															
>	> mpg %>% as.data.frame()														
	manufact	turer		mo	odel	displ	year	cyl	trans	drv	cty	hwy	fl	class	
1	L	audi			a4	1.8	1999	4	auto(15)	f	18	29	р	compact	
2	2	audi			a4	1.8	1999	4	manual(m5)	f	21	29	р	compact	
3	3	audi			a4	2.0	2008	4	manual(m6)	f	20	31	р	compact	
4	1	audi			a4	2.0	2008	4	auto(av)	f	21	30	р	compact	
5	5	audi			a4	2.8	1999	6	auto(15)	f	16	26	р	compact	
6	5	audi			a4	2.8	1999	6	manual(m5)	f	18	26	р	compact	
7	7	audi			a4	3.1	2008	6	auto(av)	f	18	27	р	compact	
5	33	ford	Α\	xplorer	4wd	5.0	1999	8	auto(l4)	4	13	17	r	suv	
	34	ford		pickup			1999	6	auto(14)	4	14	17	r	pickup	
_	35	ford		pickup			1999		manual(m5)	4	14	17	r	pickup	
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	36	ford		pickup			1999	8		4	13	16	r	pickup	
_	37	ford		pickup			1999	8	auto(14)	4	13	16	r	pickup	
8	38	ford		pickup			2008	8	auto(l4)	4	13	17	r	pickup	
8	39	ford	f150	pickup	4wd	5.4	1999	8	auto(l4)	4	11	15	r	pickup	
9	90	ford	f150	pickup	4wd	5.4	2008	8	auto(l4)	4	13	17	r	pickup	
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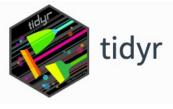
VS.

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3 audi	a4	2	<u>2</u> 008	4	manual(m6)	f	20	31	р	compac
4 audi	a4	2	<u>2</u> 008	4	auto(av)	f	21	30	р	compac
5 audi	a4	2.8	<u>1</u> 999	6	auto(15)	f	16	26	р	compac
6 audi	a4	2.8	<u>1</u> 999	6	manual(m5)	f	18	26	p	compac
7 audi	a4	3.1	<u>2</u> 008	6	auto(av)	f	18	27	р	compac
8 audi	a4 quattro	1.8	<u>1</u> 999	4	manual(m5)	4	18	26	р	compac
9 audi	a4 quattro	1.8	<u>1</u> 999	4	auto(15)	4	16	25	р	compac
l0 audi	a4 quattro	2	<u>2</u> 008	4	manual(m6)	4	20	28	р	compac



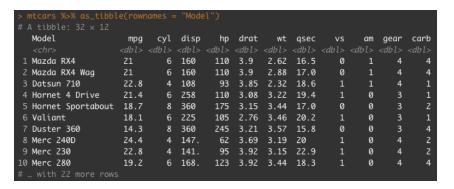
#### **Data manipulation**

- mutate() adds new variables that are functions of existing variables.
- select() picks variables based on their names.
- filter() picks rows based on their values.
- summarize () reduces multiple values down to a single summary.
- arrange () changes the ordering of the rows.
- group by () perform group-wise operations.



#### Reshaping data

- Conversion to/from Tidy data where each column is a variable and each row is an observation.
- pivot\_longer() converts to Tidy/long format.
- pivot wider() converts to wide format.
- tibble::column\_to\_rownames(var = "id\_col") converts to data frame (required for some functions).



VS.

#### + add additional variables



#### **Character string manipulations**

- str\_detect(x, pattern) looks for match to the pattern; commonly used with dplyr::filter()
- str\_extract(x, pattern) extracts the text of the match; commonly used with dplyr::mutate()
- str\_replace(x, pattern, replacement) replaces the matches with new text; commonly used with dplyr::mutate()



#### **Managing factors**

- R uses factors to handle categorical variables
- Often important to control the ordering of factors eg for plotting or modelling
- fct\_relevel() changes the order of a factor as specified by
  a character vector
- fct\_inorder() changes the order of a factor as specified by current order; commonly used with dplyr::arrange()



#### Functional programming tools for iterating with functions and vectors

- map() family of functions to replace for loops
- see the <u>Iteration</u> chapter of R for Data Science to learn more <u>https://github.com/rstudio/cheatsheets/blob/master/purrr.pdf</u>





#### Summarizes key statistical model information in tidy format

- tidy() summarizes information about model components.
- glance() reports information about the entire model.
- augment () adds information about observations to a dataset (eg residuals).
- Works with 100+ model objects.
- Plays well with the nest/unnest functions in tidyr and the map functions in purr <a href="https://broom.tidymodels.org/articles/broom\_and\_dplyr.html">https://broom.tidymodels.org/articles/broom\_and\_dplyr.html</a>



#### **Publication-quality data visualization**

- Implements a "grammar of graphics"
- Start by defining the data to be plotted ("aesthetics"):

  ggplot(aes(x, y, color, fill, shape, alpha, linetype))
- Then add layers ("geoms") to specify how data is plotted, eg:
   + geom point()
- Add additional geom layers, eg:
   + geom boxplot()
- Can split into separate plots, eg male vs. female, by "faceting":
   + facet\_wrap (~ Sex)
- Finally add title and modify theme:

```
+ labs(title = "Plot title", subtitle = "plot details"
+ theme(aspect.ratio = 1)
```

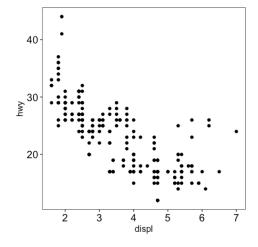


Backs

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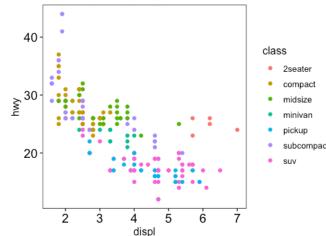


```
mpg %>%
   ggplot(aes(displ, hwy)) +
   geom_point()
```



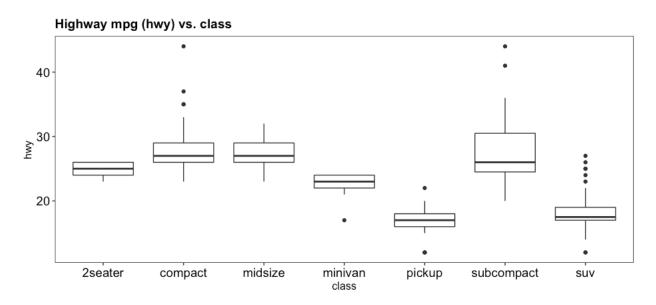
```
mpg %>%
    ggplot(aes(displ, hwy, color = class)) +
    geom_point() +
    theme(aspect.ratio = 1) +
    labs(title = "Highway mpg (hwy) vs. displacement")
```

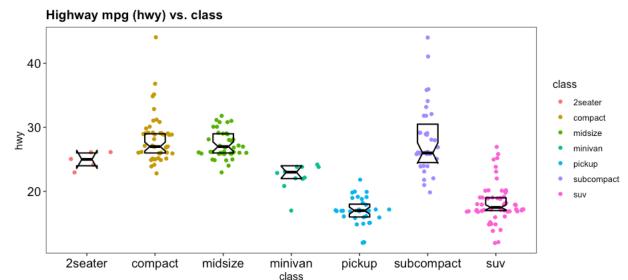
#### Highway mpg (hwy) vs. displacement





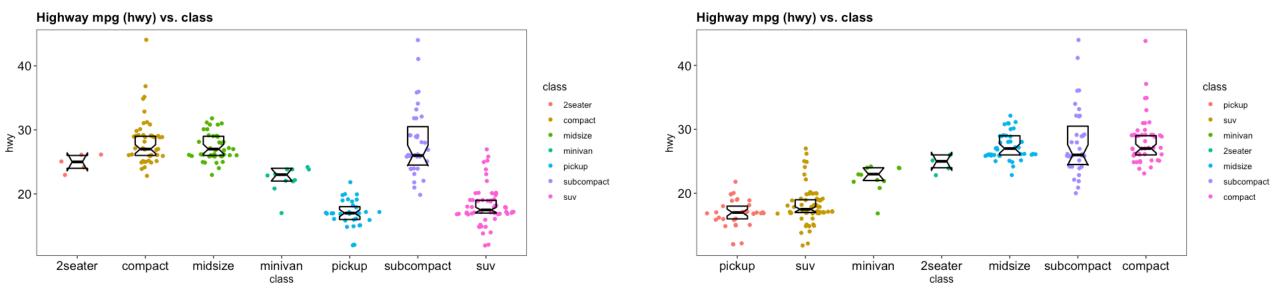
```
mpg %>%
    ggplot(aes(class, hwy)) +
    geom_boxplot() +
    labs(title = "Highway mpg (hwy) vs. class")
```



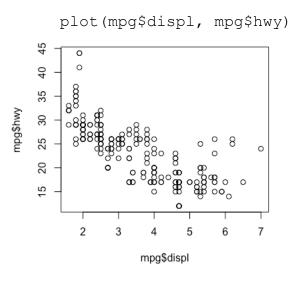


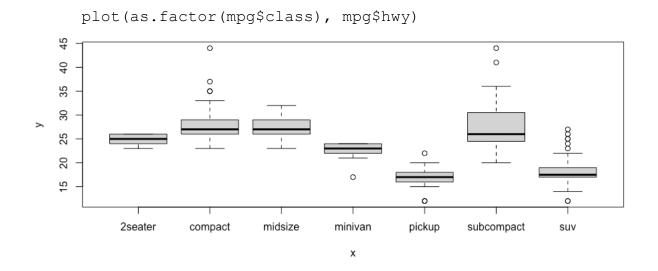


```
mpg %>%
    group_by(class) %>%
    mutate(mean = mean(hwy)) %>%
    ungroup() %>%
    arrange(mean) %>%
    mutate(class = fct_inorder(class)) %>%
    ggplot(aes(class, hwy, color = class)) +
    ggforce::geom_sina() +
    geom_boxplot(
        notch=TRUE, varwidth=FALSE, outlier.shape=NA, coef=FALSE, width=0.3, color="black", fill="transparent", size=0.75") +
    labs(title = "Highway mpg (hwy) vs. class")
```



#### Equivalent plots using base R





# Other commonly used packages

#### ggforce

Additional plot geoms, including geom\_sina() https://ggforce.data-imaginist.com/

#### ggrepel

Repel overlapping text labels <a href="https://ggrepel.slowkow.com/index.html">https://ggrepel.slowkow.com/index.html</a>

#### ggrastr

Rasterize ggplot objects or layers
<a href="https://cran.r-">https://cran.r-</a>
project.org/web/packages/ggrastr/vignettes/Raster geoms.html

#### ggsignif

Test and/or add significance brackets to plots <a href="https://cran.r-project.org/web/packages/ggsignif/vignettes/intro.html">https://cran.r-project.org/web/packages/ggsignif/vignettes/intro.html</a>

#### tidyheatmap

Tidy interface to ComplexHeatmap https://cran.r-

project.org/web/packages/tidyHeatmap/vignettes/introduction.html

#### janitor

cleaning variable names
<a href="https://cran.r-project.org/web/packages/janitor/vignettes/janitor.html">https://cran.r-project.org/web/packages/janitor/vignettes/janitor.html</a>

#### rstatix

Tidy and pipe-friendly statistics, e.g. t-test, Wilcoxon/Mann-Whitney https://cran.r-project.org/web/packages/rstatix/index.html

#### patchwork

Assemble multiple ggplot objects <a href="https://patchwork.data-imaginist.com/index.html">https://patchwork.data-imaginist.com/index.html</a>



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