Workshop:
Statistics & Bioinformatics Using R

Workshop Introduction

ASMS 2017 Annual Meeting • 6/6/2017

Ryan Benz & Jeffrey Jones

Goals of the Workshop

- Present and discuss R and the R ecosystem as a useful tool for performing scientific data analysis, visualization and reporting
- Provide a high-level view of what R can do and why it's a great language for scientific data analysis
- Show a few concrete examples of how R can be used in the context of mass spectrometry (and related) data
- Discuss the data analysis needs of the community, and how R might fit in

All workshop materials can found found at:
https://goo.gl/x247gC

Potential Questions for Discussion

- Do you see a need for a programming language tool, like R, in your research and work?
  - Are existing tools (commercial and open source) adequate for your needs?
  - Is it just easier to collaborate with statisticians/data analysts when needed?

- What are your main data challenges?
  - Existing tools don't always do exactly what I want or need
  - Data storage, handling, access, processing is hard/challenging
  - Data analysis takes too long or is difficult to do
  - …

- If there is a need for a tool like R and the need isn't being met, why not?
  - Students, employees aren't interested in using or learning
  - More training needed (academic & professional)
  - Learning to use the tools (e.g. learning to program) is too hard

More Thoughts for Discussion

- In the "–omics" age, all data is "big data"
  - Simply dealing with data can be a challenge (size, amount, complexity)
  - Standard tools (e.g. spreadsheets) are often inadequate
  - Data processing and analysis can take a *long* time, require *big* resources
- We are currently in a period of flux regarding "big data"
  - New tools, methods are constantly being developed – exciting!
  - New tools, methods are not always easy to use – frustrating!
  - What package/language/method/algorithm to use?
- For scientists, coding skills are now essential
  - We don't all have to be expert software developers
  - Basic/essential programming skills will get you a very long way
- Reproducible research is an extremely important topic in all fields of science right now – coding skills can help

Introduction to R and R Studio

R is A Great Choice for Scientists

- R describes itself as
  "a language & environment for statistical computing and graphics" - *r-project.org*

  More than just the language *Data* *Visualization*

- Free software, GPLv2 license, cross platform (Win, Mac, Linux)
- Great IDE (RStudio)
- Easy package installation and management (CRAN, Bioconductor)
- Active community
- One of the top languages for data science

Two Different Tracks for Learning R

The Programmer

1. **Python**
   learn programming fundamentals
2. **R**
   learn R fundamentals
3. **R**
   focus on data science methods

Learning is focused on forming a **strong programming base** from which to learn data science methods

The Applied Data Scientist

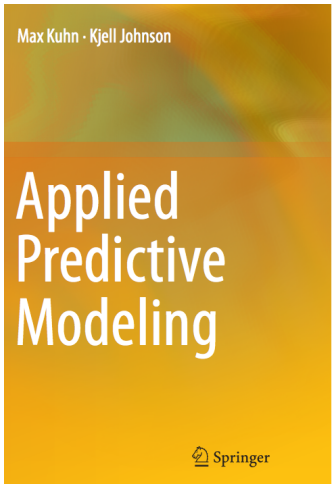
1. **R**
   learn essential data techniques
   - *file I/O*
   - *basic data munging*
   - *R data structures*
   - *plotting*
   - *basic analysis & modeling*
2. **R**
   focus on data science methods

Learning is focused on **practical coding** specifically for data analysis

R Is At The Forefront of Data Science

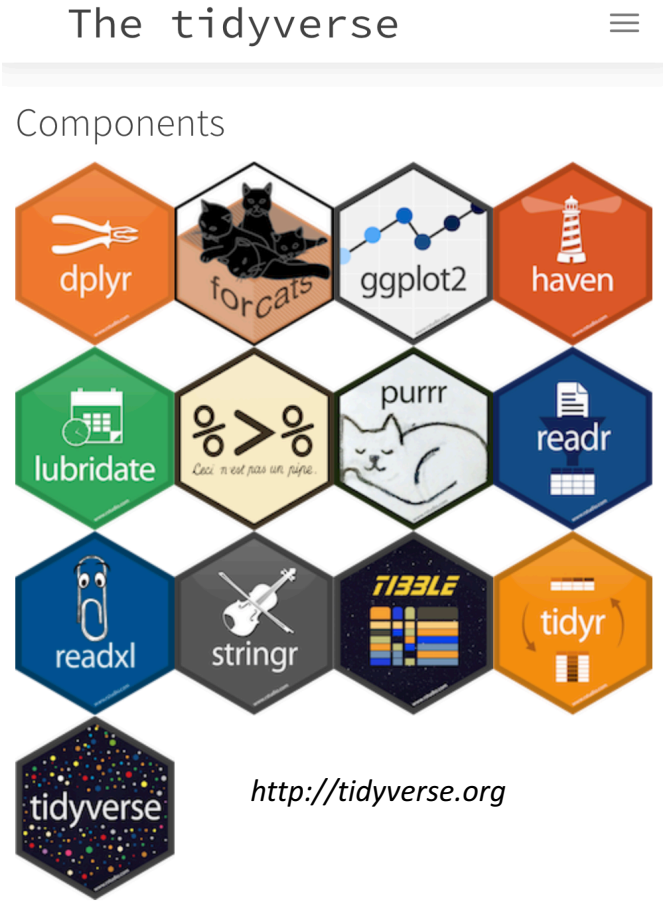
ggplot2

http://ggplot2.tidyverse.org/



http://appliedpredictivemodeling.com

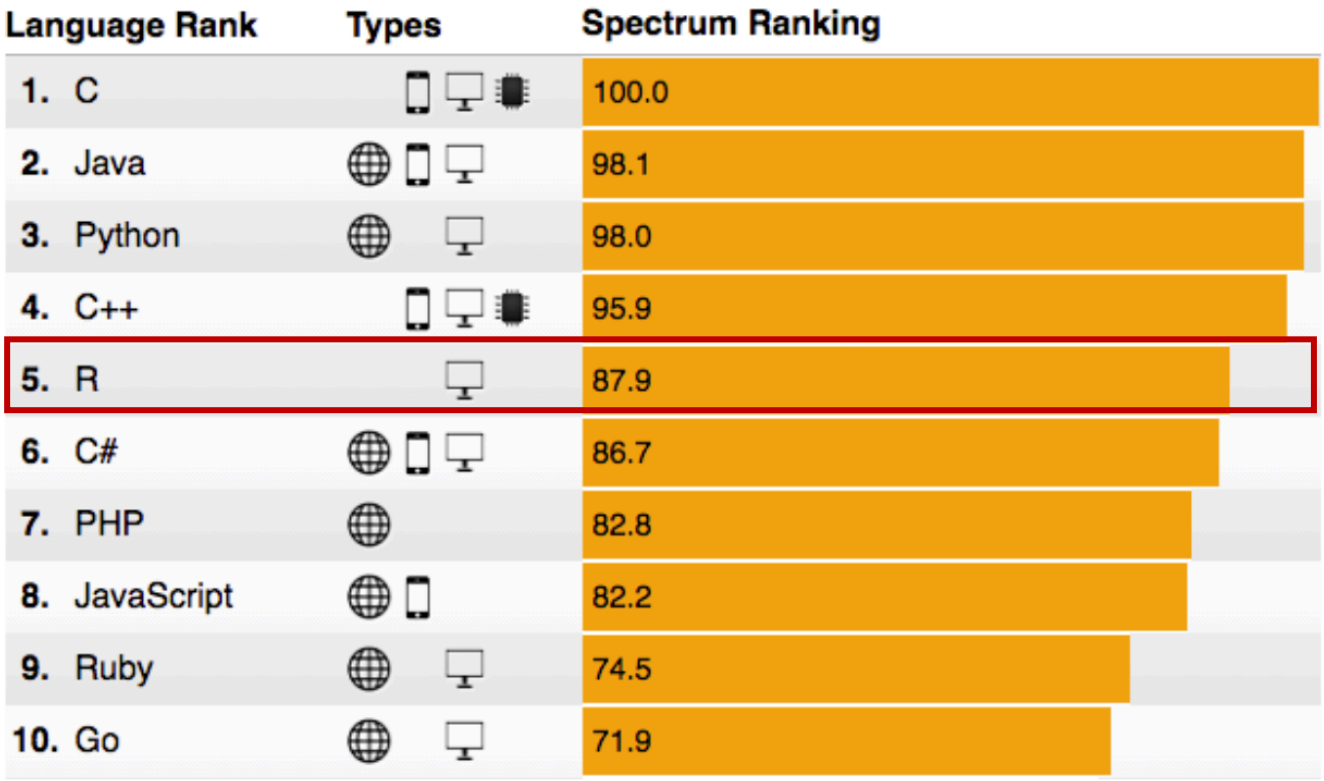
https://shiny.rstudio.com



http://tidyverse.org

R Has Become a Top Programming Language

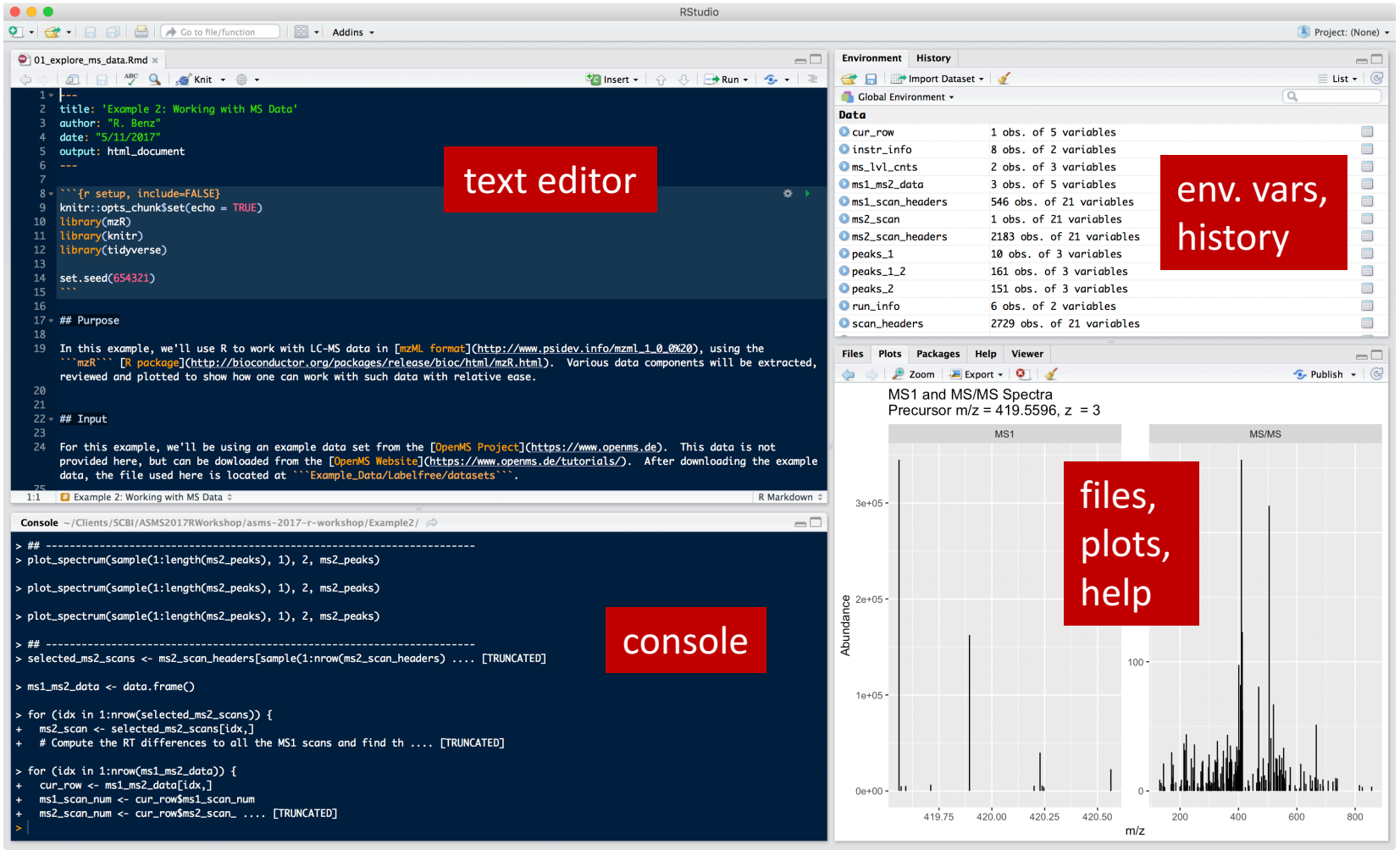
IEEE 2016 Top Programming Languages



Language Rank	Types	Spectrum Ranking
1. C		100.0
2. Java		98.1
3. Python		98.0
4. C++		95.9
5. R		87.9
6. C#		86.7
7. PHP		82.8
8. JavaScript		82.2
9. Ruby		74.5
10. Go		71.9

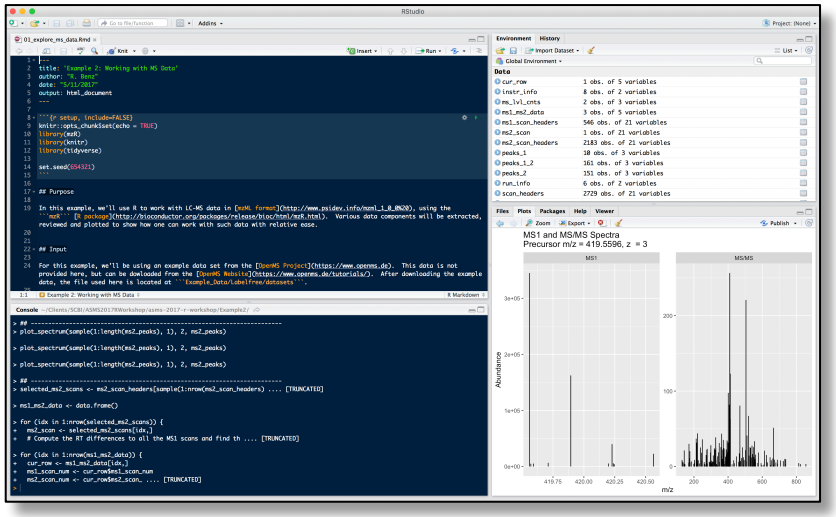
http://spectrum.ieee.org

RStudio:
A Top Notch IDE for Data Analysis

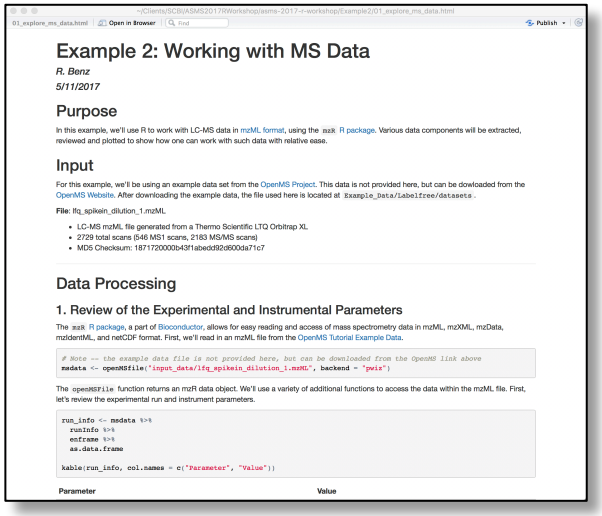


RStudio Does More Than Just Code

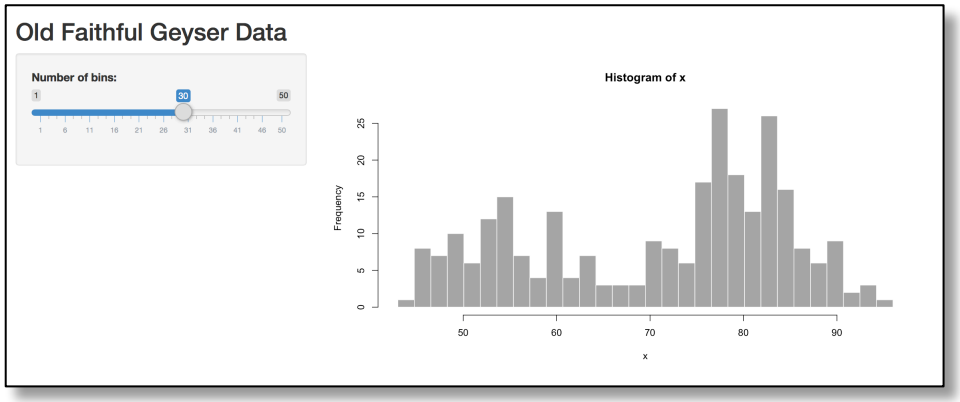
Data analysis workspace



Reproducible Reports



Interactive Web Applications (Shiny)



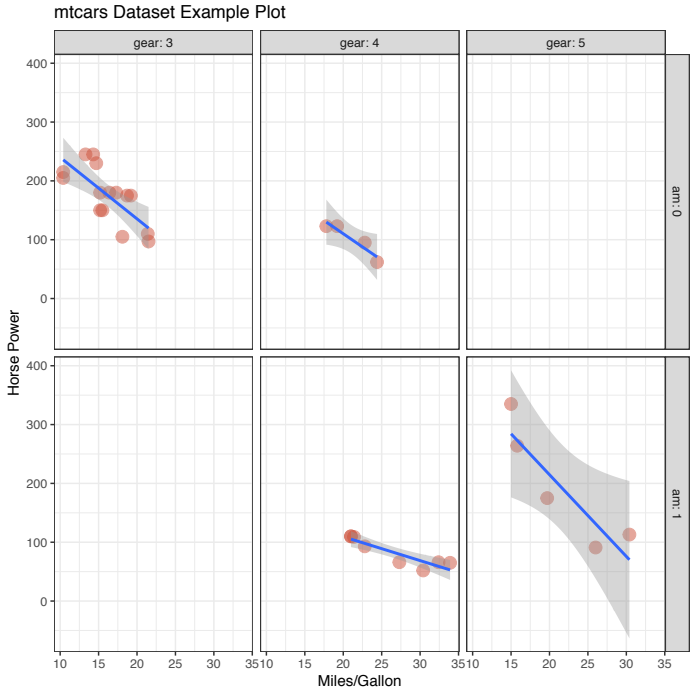
- Git/GitHub integration
- Publish to the web
- Work in the cloud with RStudio Server

A ggplot2 Example

The Code

```
# Set-up the plot & specify data
ggplot(mtcars, aes(mpg, hp)) +
  # Represent data as points
  geom_point(size = 4, color = "tomato3", alpha = 0.5) +
  # Separate out the data by transition type and # of gears
  facet_grid(am ~ gear, labeller = label_both) +
  # Add a linear model fit for each group
  geom_smooth(method = "lm") +
  # Label the x-axis
  xlab("Miles/Gallon") +
  # Label the y-axis
  ylab("Horse Power") +
  # Add a title
  ggtitle("mtcars Dataset Example Plot") +
  # Use the "black and white" theme
  theme_bw()
```

The Output

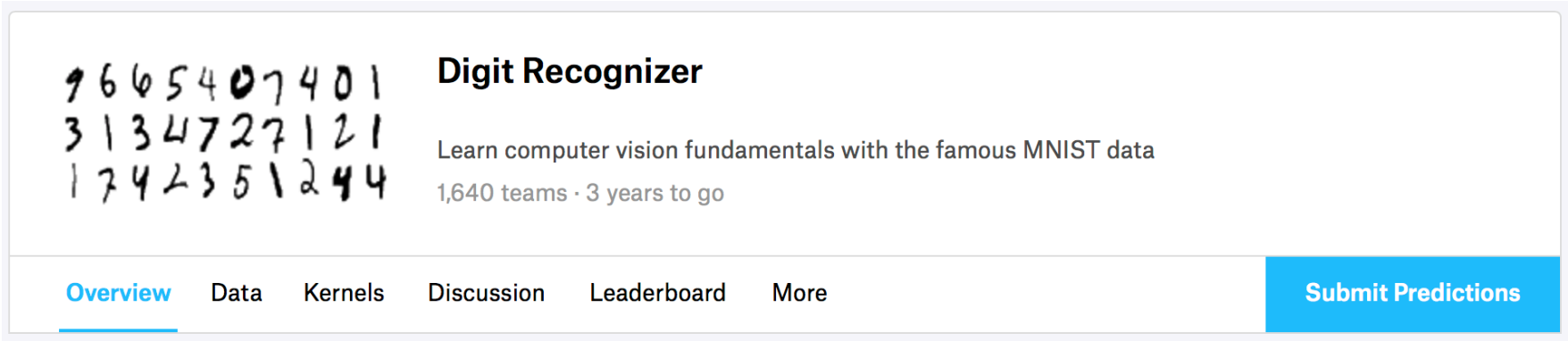


Plots are built-up layer by layer,
giving you the flexibility to create just what you

A Predictive Modeling Example

The Task

Given an image of a handwritten digit, predict what the digit actually is



Data, example code, and competition leaderboards all available from Kaggle (*https://www.kaggle.com/c/digit-recognizer*)

A Predictive Modeling Example

Code

This script has been released under the Apache 2.0 open source license.

Download Code

```
# Creates a simple random forest benchmark

library(randomForest)
library(readr)

set.seed(0)

numTrain <- 10000
numTrees <- 25

train <- read_csv("../input/train.csv")
test <- read_csv("../input/test.csv")

rows <- sample(1:nrow(train), numTrain)
labels <- as.factor(train[rows,1])
train <- train[rows,-1]

rf <- randomForest(train, labels, xtest=test, ntree=numTrees)
predictions <- data.frame(ImageId=1:nrow(test), Label=levels(labels)[rf$test$predicted])
head(predictions)

write_csv(predictions, "rf_benchmark.csv")
```

This model is 93.5% accurate on the test set

Most of the code is about preparing the data

Build the model and make predictions in 2 lines of code!

https://www.kaggle.com/benhamner/random-forest-benchmark-1/code/code

dplyr Data Pipelines

- Makes common data analysis manipulations easy and human readable

- Pipelines are built from
  data sets + data manipulation verbs (functions)

  - `mutate()` adds new variables that are functions of existing variables
  - `select()` picks variables based on their names.
  - `filter()` picks cases based on their values.
  - `summarise()` reduces multiple values down to a single summary.
  - `arrange()` changes the ordering of the rows.

dplyr Data Pipelines

Consider the mtcars data set

```
> head(mtcars)
                   mpg cyl disp  hp drat    wt  qsec vs am gear carb
Mazda RX4         21.0   6  160 110 3.90 2.620 16.46  0  1    4    4
Mazda RX4 Wag     21.0   6  160 110 3.90 2.875 17.02  0  1    4    4
Datsun 710        22.8   4  108  93 3.85 2.320 18.61  1  1    4    1
Hornet 4 Drive    21.4   6  258 110 3.08 3.215 19.44  1  0    3    1
Hornet Sportabout 18.7   8  360 175 3.15 3.440 17.02  0  0    3    2
Valiant           18.1   6  225 105 2.76 3.460 20.22  1  0    3    1
```

… 32 total entries

dplyr Data Pipelines

Goal
Compute the mean horsepower
by number of cylinders

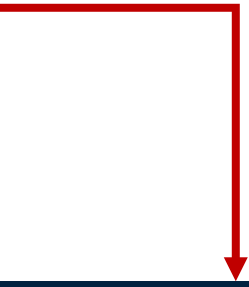
A simple first approach might
look like this

```
hp_vals_4 <- c()
hp_vals_6 <- c()
hp_vals_8 <- c()
for (i in 1:nrow(mtcars)) {
  current_car <- mtcars[i,]

  cyl <- current_car$cyl
  hp <- current_car$hp

  if (cyl == 4) {
    #... accumulate values
    hp_vals_4 <- c(hp_vals_4, hp)
  } else if (cyl == 6) {
    #... accumulate values
  } #...
  #...
}
# compute summary stats
mean_hp_4 <- mean(hp_vals_4)
#...
```

dplyr Data Pipelines

The dplyr approach is MUCH simpler, easier to read and understand (and does more)

```
mtcars %>%
  group_by(cyl) %>%
  summarize(n = n(),
            mean_hp = mean(hp),
            median_hp = median(hp),
            stdev_hp = sd(hp))
```



```
# A tibble: 3 x 5
    cyl     n   mean_hp median_hp stdev_hp
  <dbl> <int>     <dbl>     <dbl>    <dbl>
1     4    11  82.63636      91.0 20.93453
2     6     7 122.28571     110.0 24.26049
3     8    14 209.21429     192.5 50.97689
```

dplyr Data Pipeline

Examples of other questions you can easily "ask"

Which automatic car has the most horsepower?

```
mtcars %>%
  filter(am == 0) %>%
  filter(hp == max(hp))
```

Which automatic and manual cars are the most fuel efficient?

```
mtcars %>%
  group_by(am) %>%
  filter(mpg == max(mpg)) %>%
  arrange(desc(mpg))
```

dplyr allows you to focus on the question not the implementation details of how to answer it

A Shiny Example

ui.R

```
library(shiny)

shinyUI(fluidPage(

  # Application title
  titlePanel("Old Faithful Geyser Data"),

  # Sidebar with a slider input for number of bins
  sidebarLayout(
    sidebarPanel(
      sliderInput("bins",
                  "Number of bins:",
                  min = 1,
                  max = 50,
                  value = 30)
    ),

    # Show a plot of the generated distribution
    mainPanel(
      plotOutput("distPlot")
    )
  )
))
```

Define the interface

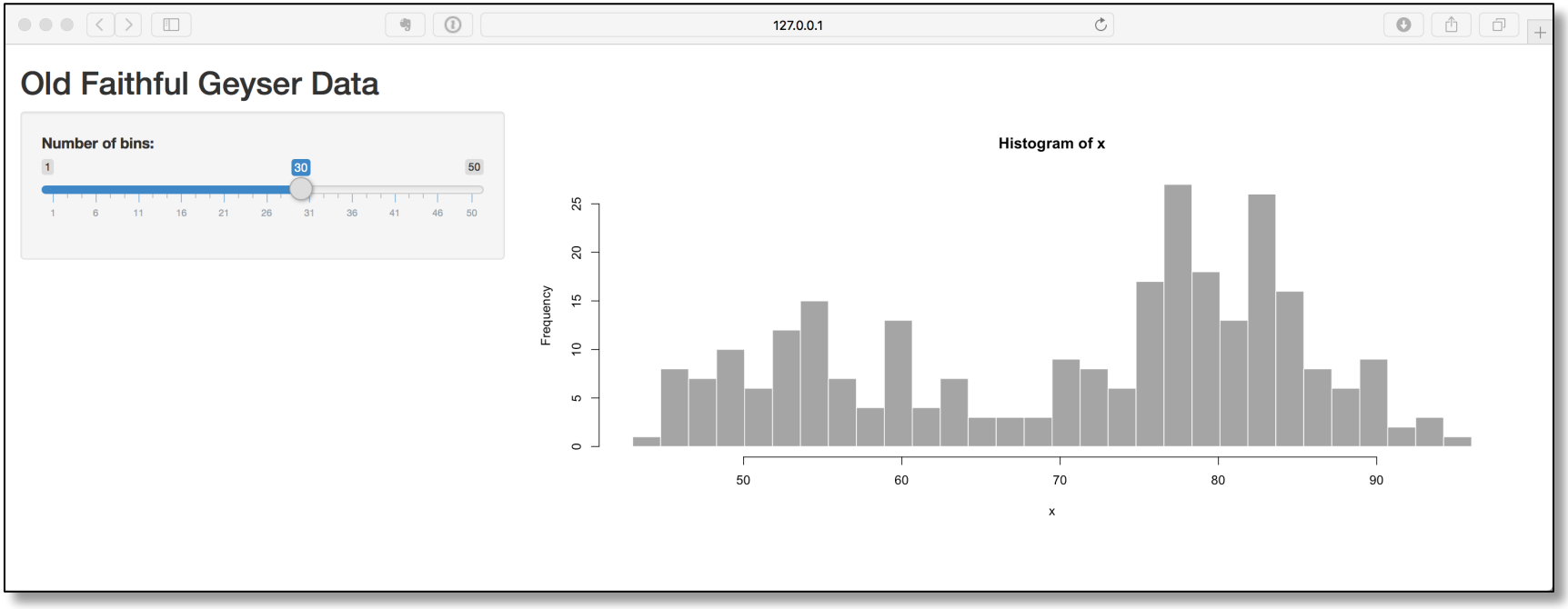
server.R

```
library(shiny)

shinyServer(function(input, output) {

  output$distPlot <- renderPlot({

    # generate bins based on input$bins from ui.R
    x    <- faithful[, 2]
    bins <- seq(min(x), max(x), length.out = input$bins + 1)

    # draw the histogram with the specified number of bins
    hist(x, breaks = bins, col = 'darkgray', border = 'white')

  })

})
```

Define the application logic, functionality

Default Shiny example code
https://www.rstudio.com

A Shiny Example



A full, interactive (albeit very simple) web app created in <50 lines of code
No need to understand any web technologies

Default Shiny example code
https://www.rstudio.com

Getting Started with R & RStudio

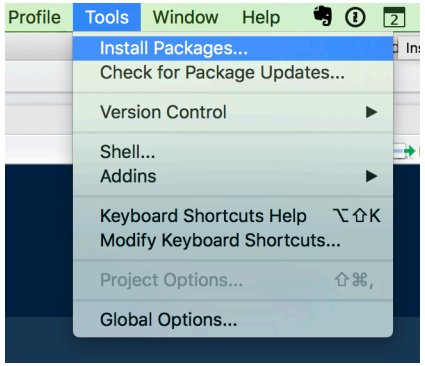
Follow these steps to run and work with the workshop examples

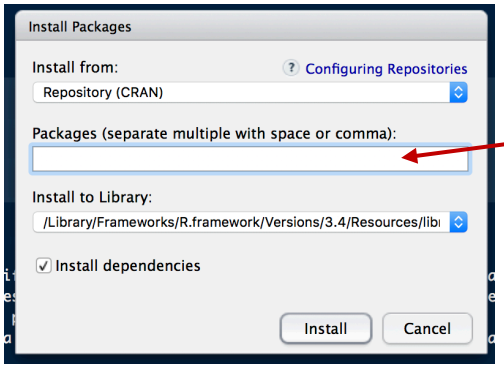
1. Download and install R
   https://www.r-project.org
   - Choose the cloud.r-project.org mirror, https is preferable
   - Latest version as of 06/02/2017:
     3.4.0 (2017-04-21) -- "You Stupid Darkness"

2. Download and install RStudio
   https://www.rstudio.com/products/rstudio/
   - Choose the Desktop, Open Source Edition (free)

Getting Started with R & RStudio

3. Install R packages from RStudio





Package names
tidyverse
knitr

4. Install mzR from Bioconductor from the R prompt
https://bioconductor.org/packages/release/bioc/html/mzR.html

Installation

To install this package, start R and enter:

```
## try http:// if https:// URLs are not supported
source("https://bioconductor.org/biocLite.R")
biocLite("mzR")
```

Additional Topics & Examples

- R Example #1: Working with Experimental Meta-data
- R Example #2: Workings with LC-MS Data
- Example interactive web application (Shiny)
- Discussion of key data science themes for scientific analysis: Reproducible data analysis

Key Data Science Themes for Scientific Data Analysis

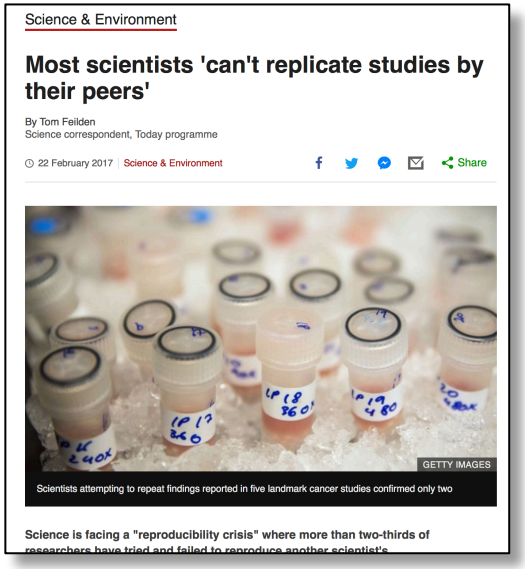
The "Reproducibility/Replication Crisis" is An Important Current Topic

Essay

Why Most Published Research Findings Are False

John P. A. Ioannidis

PLoS Medicine, Aug 2005 2(8)



Science & Environment

Most scientists 'can't replicate studies by their peers'

By Tom Feilden
Science correspondent, Today programme

22 February 2017 | Science & Environment

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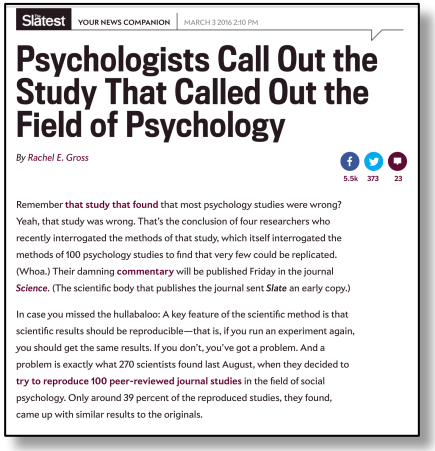
Scientists attempting to repeat findings reported in five landmark cancer studies confirmed only two

Science is facing a "reproducibility crisis" where more than two-thirds of researchers have tried and failed to reproduce another scientist's



Slatest YOUR NEWS COMPANION MARCH 3 2016 2:10 PM

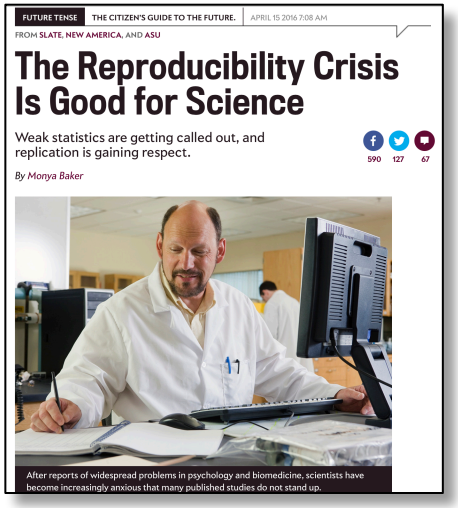
Psychologists Call Out the Study That Called Out the Field of Psychology

By Rachel E. Gross

5.5k 373 23

Remember **that study that found** that most psychology studies were wrong? Yeah, that study was wrong. That's the conclusion of four researchers who recently interrogated the methods of that study, which itself interrogated the methods of 100 psychology studies to find that very few could be replicated. (Whoa.) Their damning **commentary** will be published Friday in the journal ***Science***. (The scientific body that publishes the journal sent ***Slate*** an early copy.)

In case you missed the hullabaloo: A key feature of the scientific method is that scientific results should be reproducible—that is, if you run an experiment again, you should get the same results. If you don't, you've got a problem. And a problem is exactly what 270 scientists found last August, when they decided to **try to reproduce 100 peer-reviewed journal studies** in the field of social psychology. Only around 39 percent of the reproduced studies, they found, came up with similar results to the originals.



FUTURE TENSE THE CITIZEN'S GUIDE TO THE FUTURE. APRIL 15 2016 7:08 AM
FROM SLATE, NEW AMERICA, AND ASU

The Reproducibility Crisis Is Good for Science

Weak statistics are getting called out, and replication is gaining respect.

590 127 67

By Monya Baker

After reports of widespread problems in psychology and biomedicine, scientists have become increasingly anxious that many published studies do not stand up.

http://www.slate.com/blogs/the_slatest/2016/03/03/psychology_study_that_induced_the_reproducibility_crisis_was_wrong.html
http://www.bbc.com/news/science-environment-39054778
http://www.slate.com/articles/technology/future_tense/2016/04/the_reproducibility_crisis_is_good_for_science.html

The Reproducible Data Analysis Problem Can Be Addressed By Software Dev/Data Science Best Practices

Some examples include…

Tidy Data

- Organizing (raw) data into a well structured form ready for data analysis
- Tidy data facilitates straight-forward data analysis and sharing with others
- The steps performed to tidy data need to be well documented

Data Analysis Pipelines

- All analysis steps should be captured in a set of scripts or programs
- Analysis steps, input, processes should be well documented
- Anyone should be able to reproduce the complete analysis path

Literate Programming

- A programming paradigm where code and documentation live in the same "document", human-centered
- A natural fit for scientific reporting
- Analysis code lives side-by-side with the results

Version Control Systems

- Central repositories for storing and tracking code changes
- Helps manage code, avoid incorrect versions and conflicts
- Facilitates open sharing of code (e.g. GitHub)

A Data Analysis Might Not Be Reproducible If…

- you had to copy and paste at some point
- you had to manually edit the data, in any way
- you had to touch a mouse to perform any part of the analysis
- an outside person can't reproduce the analysis

Some additional complex questions to consider:

- Are your platform, operating system, software versions reproducible?
- Are your inputs (e.g. data and parameters) reproducible?
- Could "future you" reproduce your own analysis?

A Few Thoughts

- Reproducibility does not imply correctness/accuracy
  - you can still do the wrong thing in a reproducible way!
  - …but knowing EXACTLY what went wrong is an important first step to correcting the mistake
  - sharing your code with others can help you find problems you didn't even know about

- There's a continuum of complexity for doing reproducible data analysis, lots of factors to consider

- At the very least:
  strive to capture all analysis steps in as set of programs/scripts!

Resources

Learning R

- Learning R and Data Science Methods
  - Coursera online classes
    e.g. JHU Data Science Series
    https://www.coursera.org/specializations/jhu-data-science
  - RStudio's curated learning resource lists
    https://www.rstudio.com/online-learning/
- Books
  - R for Data Science: http://r4ds.had.co.nz
  - ggplot2: Elegant Graphics for Data Analysis, 2nd Ed.
  - Advanced R: http://adv-r.had.co.nz
- Practice
  - UCI Machine Learning Repo: http://archive.ics.uci.edu/ml/
  - Kaggle Datasets & Competitions: https://www.kaggle.com
  - Build a small tool that make one part of your job easier

Additional Materials

- Books
  - *Applied Predictive Modeling* (Kuhn, Johnson)
  - *The Grammar of Graphics* (Wilkinson)
- Websites
  - R Bloggers (https://www.r-bloggers.com)
  - Kaggle (http://www.kaggle.com)
  - FlowingData (http://flowingdata.com)
- Articles
  - Tidy Data (http://vita.had.co.nz/papers/tidy-data.html)
  - A Layered Grammar of Graphics (http://vita.had.co.nz/papers/layered-grammar.html)
- Visualization Frameworks
  - d3.js (https://d3js.org)
  - Processing, p5.js (https://processing.org)