R bootcamp - part 2: visualisation with ggplot2

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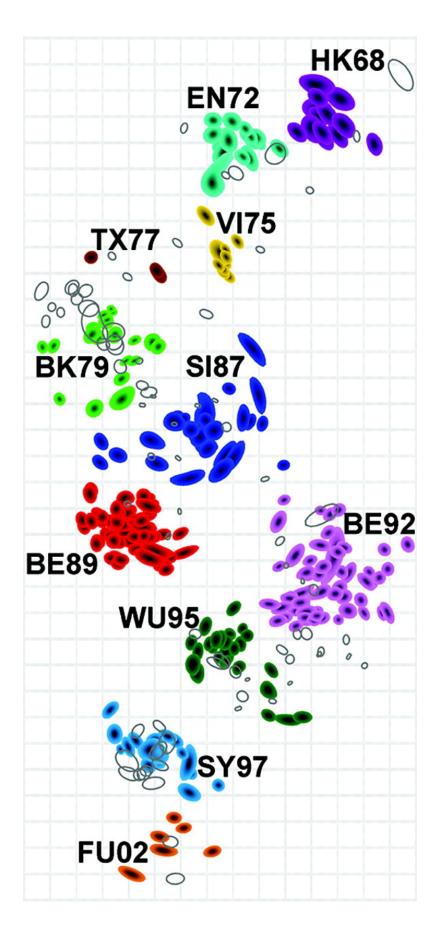
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1 Background: Antigenic cartography

Antigenic Cartography is the process of creating maps that reflect the antigenic properties of a pathogen. In this course, we will use antigenic cartography data used to visualise and quantify the antigenic evolution of influenza viruses.

To create antigenic maps, measurements of laboratory binding assays are converted to a coordinate space in which the distance between points reflects their similarity in the original assay. For instance, to create an antigenic map of influenza viruses, viruses can be titrated against sera in a hemagglutination inhibition (HI) assay. The HI assay tests the ability of influenza viruses to agglutinate red blood cells and the ability of antisera raised against the same or related strains to block this agglutination. Sera with similar abilites to block agglutination will be close together in the antigenic map and simiarly, antigens with similar titers will be nearby in antigenic space.

Smith et al (2004) mapped the antigenic evolution of human influenza A (H3N2) viruses, which have been a major cause of influenza epidemics since the 1968 Hong Kong influenza pandemic Smith et al. (2004). We will use their antigenic coordinates of viruses that circulated world-wide circulation between 1968 and 2003, which generated the following map:



At the end of this course, you will be able to recreate this map, plot the distribution of measurement counts over time and visualise the location that viruses were isolated on a word map.

2 Setting up

2.1 Starting an Rproject for your analysis

First, we will set up a project, where you will keep all files associated with that project - including your analysis reports, input data, results and figures.

- In R studio click File > New Project.
- Choose Existing directory, then click on Browse to find the folder that you created with the course material
- Click Create Project

In your files plane in R studio you can now see all the files you downloaded for this workshop. We will be able to easily read them into R and save results there.

Note: There are other ways of specifying the folders from which you read and where you write results and save plots to, but I would highly recommend this strategy. It keeps everything neat and you have all the important parts of your analysis in one place.

When you start your own analysis (independent of this tutorial), follow the scheme above:

- create a projects in R
- choose Existing directory, then click on Browse to find the folder with your data
- Click Create Project

To generate a new file for you analysis:

- choose File > New File
- select:
 - R Markdown for a file like the one you are looking at i.e. with text and code chunks interleaved
 - R Script for a file that only contains plain code, i.e. no need for chunks as the entire file is one big chunk!

2.2 Starting an Rproject for your own analysis: some additional advice

- Have a look at this blog post, with more advice on the structure and sub-folders in your project directory.
- Rstudio provides a number of cheat sheets for the RStudio interface setup, R Markdown etc. To acces them, choose Help > Cheatsheets:
 - RStudio IDE Cheat Sheet
 - R Markdown Cheat Sheet

2.3 Customising RStudio settings

RStudio can be customised and I leave this to everyone to figure out what works best. However, I would recommend changing one default setting, which will ensure that when you start working in your project, you start of with a clean slate and none of previously computed data sticks around and confuses your analysis.

- on Mac OS:
 - go to RStudio > Preferences

- in General untick restore .RData into workspace at startup.
- on Windows and Linux:
 - go to RStudio > Tools
 - in global options untick restore .RData into workspace at startup.

We are all set now to go ahead with your first analysis and data visualisation in R!

2.4 Setting up an analysis report

The document you are looking at right now is a R Notebook. R Notebooks allow us to interleaf text describing our analysis with the R code that actually contains the analyses commands.

The text follows some simple markdown rules (for instance bold header sections etc, which we will not go into detail here). Important for us at this stage is that whenever we want to include analysis code into the document, we have to create an R code chunk. To include a new chunk click the Insert button at the top of your editor window and select R.

All code *chunks* have some default settings, concerning their layout, execution etc, which can be heavily customised. For our beginners tutorial, we do not have to worry about all of these. I mention this here, as the following and first chunk of our document contains some basic options that I want to have applied to all chunks in the rest of the document. Specifically, it tells R studio that when I prepare this document for sharing with you as a pdf, that I want both the actual code and the results displayed in the document. It also specifies the width and alignment of the figures in the final document.

I then follow with a chunk that loads all libraries required for my analysis. The following chunk loads the libraries that you installed as a preparation for the course into your R workspace:

```
library("tidyverse")
library("sf")
library("rnaturalearth")
library("RColorBrewer")
```

Note: Libraries only have to be installed once, however, they will have to be loaded into the R workspace whenever we open a new R session. Think of the libraries you install as tools that you buy: you buy a hammer the first time you realise you want to hang a picture and need a hook in the wall. Once you bought it, you need to actually bring it to the room where you want to hang the picture. After that, you have the hammer and can use it whenever you like. The hammer is the library that you install (buy) once and then load into your R workspace (use) whenever you have a task that can be accomplished with that hammer.

3 Data input

The first step in data analysis with R is to read the data into the R workspace. We can do this with the read_csv function.

3.1 A primer to R functions

Functions automate common tasks such as reading files into R, creating a histogram of your data and saving that histogram in a pdf document.

Functions take a set of arguments, evaluate them and return the result. There are two possible outcomes that we might want to see when we use a function:

If we simply want to see the result of the function displayed after execution, we just type the command and execute it. If we want to store the result in a variable for future use, we have to assign the outcome of a function into an object by using the assign operator <-.

To see a description of the function, its arguments and results, use the help function? by typing <code>?function_name</code> in the R console. This help function proves really useful whenever you want to use a new function or you want to look up some examples of how to use the function.

4 Reading your data

For this tutorial, I have downloaded the data from Smith *et al* (2004) (made available at the following http://www.antigenic-cartography.org/)) and formated for us to work with. In the following chunk, we pass the filename of this formated data set, located in the data folder of your project, as the argument to the read_csv function and assign the ouput to a new object called coord.

```
coord <- read_csv("data/2004_Science_Smith_data.csv")
#> Rows: 322 Columns: 9
#> -- Column specification ------
#> Delimiter: ","
#> chr (4): name, cluster, type, location
#> dbl (5): year, x.coordinate, y.coordinate, lat, lng
#>
#> 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.
```

read_csv() prints out a column specification that gives the name and type of each column.

4.1 Exercises

- 1. Use the help function? to have a look at the documentation of read csv.
- 2. Have a look at the coord object by creating a new chunk, typing coord and executing the code chunk.
- 3. Do you find the message printed by read_csv represented in coord?

```
coord
#> # A tibble: 322 x 9
#>
                                       x.coordinate\ y.coordinate\ location
      n.a.me.
                   year cluster type
                                                                              lat
                                                                                    lnq
#>
      <chr>
                  <dbl> <chr>
                                 <chr>
                                              <dbl>
                                                            <dbl> <chr>
                                                                            <db1>
                                                                                 <dbl>
#>
    1 BI/15793/~
                   1968 HK68
                                AG
                                               4.05
                                                             15.0 BILTHOV~
                                                                            52.1
                                                                                   5.02
    2 BI/16190/~
                   1968 HK68
                                               4.10
                                                             14.8 BILTHOV~
                                                                            52.1
                                AG
    3 BI/16398/~
                   1968 HK68
                                AG
                                                             13.9 BILTHOV~
                                                                            52.1
                                                                                   5.02
#>
                                               4.36
                   1969 HK68
                                               3.87
#>
    4 BI/808/69
                                AG
                                                             14.3 BILTHOV~
                                                                            52.1
   5 BI/908/69
#>
                   1969 HK68
                                AG
                                               4.87
                                                             14.1 BILTHOV~
                                                                            52.1
                                                                                   5.02
#>
    6 BI/17938/~
                   1969 HK68
                                AG
                                               4.40
                                                             14.9 BILTHOV~
                                                                            52.1
                                                                                   5.02
#>
  7 BI/93/70
                                                                             52.1
                                                                                   5.02
                   1970 HK68
                                AG
                                               5.06
                                                             14.5 BILTHOV~
   8 BI/2668/70
                  1970 HK68
                                AG
                                               4.82
                                                             15.5 BILTHOV~
                                                                             52.1
                                                                                   5.02
  9 BI/6449/71
                                                             15.9 BILTHOV~
                                                                            52.1
                                                                                   5.02
                 1971 HK68
                                AG
                                               3.87
```

5 Data and object types

The objective of this course is data visualisation and we will not spend a huge amount of time on learning R's basic obeject structures. Instead, we will focus on the functions and data objects from R tidyverse. The tidyverse is an 'opinionated collection of R packages designed for data science. All packages share an underlying design philosophy, grammar, and data structures. While there are many other options for wrangling your data, formating and plotting, I would suggest to learn this from the beginning as it is very neat and makes the code very readable.

5.1Data types

The most common data types in R (base R and tidyverse) are:

- int, which stands for integers i.e 1, 2, 3;
- dbl, which stands for doubles, or real numbers i.e. 1.2, 1.7, 9.0;
- chr, which stands for character vectors, or strings i.e. "a", "b", "word";
- lgl, which stands for logical, vectors that contain only TRUE or FALSE;
- fctr, which stands for factors, which R uses to represent categorical variables with fixed possible values.

5.2 Object types

There are many different data types in R. For the purpose of data visualisation and this workshop, we will only work with two object types:

- data.frame: a list of variables with the same number of observations. Variables are in columns, observations in rows. Rows have unique rownames. data.frames are one common data structure in base
- tibble: tibbles are 'opinionated data frames from the tidyverse' with an improved printing display, stricter rules for re-formating that aid in avoiding bugs and no rownames.
- c(): one dimensional vector with its elements separated by commas, e.g c(1.6, 2.5, 3.2) is a vector with three elements of the type double.

Note: Tibbles are newer than data frames and some old functions will not work with them and might require actual data frames; For the majority of our analyses and visualisations we will work with tibbles.

5.3 Exercises:

Use the output of running coord to determine:

- 1. which data types are present in our dataset;
- 2. how many observations and variables are in our dataset.

Available data from antigenic cartography study 6

I processed and formated the publically available data from Smith et al. (2004) to generate the data you know see when running coord. The variable names (i.e. column names) specify what the data is we are looking at:

name	name of virus isolate
year	year of isolation
cluster	derived cluster
type	serum or antigen measurement
x.coordinate	x coordinate in antigenic space
y.coordinate	y coordinate in antigenic space
location	location of virus measurement
lat	latitude of location
lng	longitude of location

7 A recipe for generating graphs with ggplot2

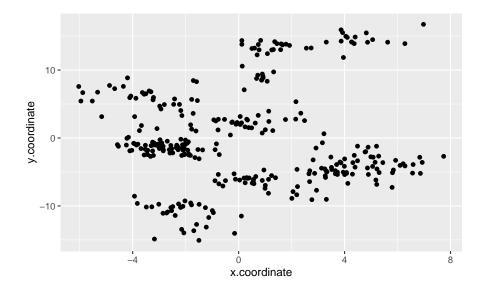
In the following section, we will generate our first plots using the ggplot2 package. Data visualisation in ggplot2, however simple or complex, follows a general recipe:

- 1. Setting up a coordinate system with the function ggplot(): provide the dataset to use in the graph
- 2. Adding layers with geom_xxx() functions:
- layers are quite literally added to ggplot() object, by using the + operator;
- each geom function expects a mapping argument which defines how variables are mapped to visualisation. The mapping argument is provided with aes(), where the x and y arguments describe which variables to map to the x and y axes. ggplot2 looks for the mapped variables in the data argument to ggplot();
- There are many geom functions that each add a different type of layer to a plot. Their names are very descriptive, for instance:
 - geom_point adds a layer of points to the coordinate system, effectively creating a scatterplot;
 - geom_histogram adds a histogram layer;
 - geom_boxplot adds a boxplot layer.

Note: The add operator + can never be at the start of a line! When adding multiple layers to a plot, we will always end the layer line with the + sign, never start a new layer with a +.

7.1 Our first plot

```
p <- ggplot(data=coord)
p + geom_point(mapping=aes(x=x.coordinate, y=y.coordinate))</pre>
```



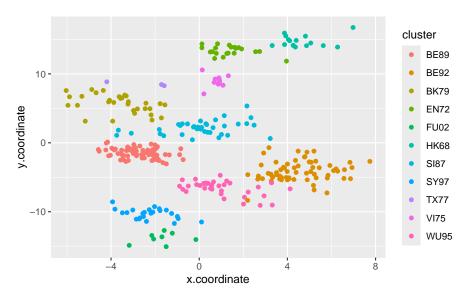
7.2 Exercises

- 1. Run ggplot(data = coord). What do you see?
- 2. What makes this simple plot look very different from the map that we want to achieve?
- 3. What other information in our data object coord could we use?

7.3 Mapping additional aesthetics

To map additional information onto our 2d scatter plot, ggplot2 makes use of aesthetics. We have already seen aesthetics in the example above, where we mapped the x.coordinate and y.coordinate to the x- and y-axis using aes(x=x.coordinate, y=y.coordinate). Broadly speaking, aesthetics are the visual properties of the objects in your plot. They include for instance the size, shape, or color of your points. The different flavors of an aesthetic are called levels. The levels in the shape aesthetic are for instance round, triangular and square. Levels of the color aesthetic could be blue, red and yellow. In our graph above, we have not used any of these aesthetics yet. Let's start by introducing color to the plot. As in the original publication, we can color the points in our plot by cluster name. We do this by simpling specifying the color aestetic in the mapping:

```
p + geom_point(aes(x=x.coordinate, y=y.coordinate, color=cluster))
```



ggplot2 automatically assign a unique color level to each unique value of cluster. This assignment process is called scaling. Depending on data and aesthetic, ggplot2 selects a reasonable scale and constructs a legend that explains the mapping between levels and variable values, in this case color and cluster. However, we can also provide our own color-scheme.

7.4 Setting scales

Adding scales to ggplot objects follows the same scheme as adding layers: we add the scale to the existing object by the + operator. Similar to geom_xxx, we have scale_xxx_yyy: xxx specifies the aestetic for which we are providing the scale, yyy specifies the type of scale we want. For instance:

- scale_color_continuous sets a continuous scale for the asthetic color;
- scale_shape_discrete sets a discrete scale for the asthetic shape.

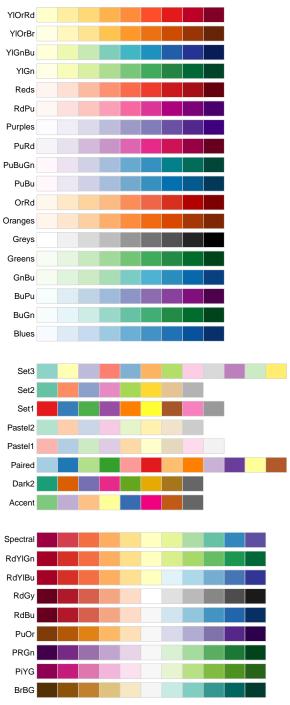
When selecting a scale, we need to consider what type of data we are displaying and what message we want to convey:

- qualitative data: unordered, distinct categories, as in our example cluster names;
- sequential data: ordered data that progresses from low to high, as in our example 'year of isolation';
- diverging data: data from low to high, with emphasis on mid-range values as well for instance correlations that range from -1 to 1, where the mid-range around 0 ie no correlation are equally important to be visualised

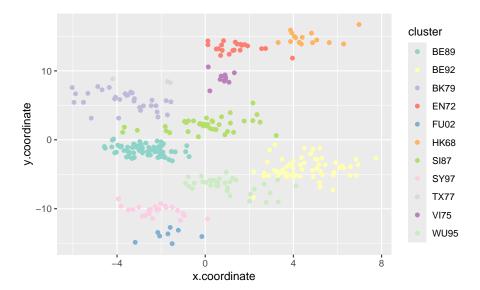
7.4.1 Color scales

The colorbrewer website provides a great resource to pick appropriate color scales.

ggplot2 has direct access to these color schemes:



```
p +
    geom_point(aes(x=x.coordinate, y=y.coordinate, color=cluster)) +
    scale_color_brewer(type="qual", palette = "Set3")
```



7.4.2 Shape scales

There are 25 point shapes available in R:

0	1	<u>2</u>	3	4 ×	
5	6	7	8	9	
10 ⊕	11 💢	12 ⊞	13 ⊠	14	
15	16	17	18 ◆	19	
20	21	22 	23	24	25

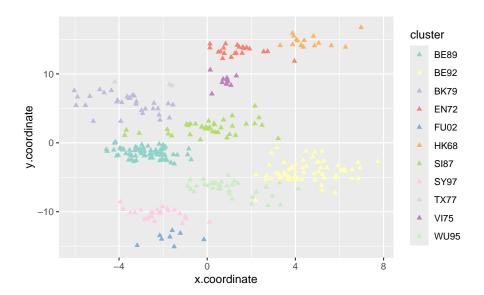
We can use the shape aesthetic and scale in analogy to how we specified color.

Note: Shapes 0-20 work in conjunction with the color aestetic, shapes 21-25 with the color and fill aesthetic.

7.5 Manual aesthetics

In addition, we could also decide that we would like to display all our points in the data set as number 17 triangles. To set an aesthetic manually, you move it outside the mapping argument and specify the level:

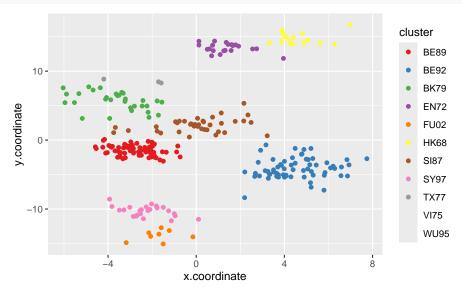
```
p +
   geom_point(aes(x=x.coordinate, y=y.coordinate, color=cluster), shape=17) +
   scale_color_brewer(type="qual", palette = "Set3")
```



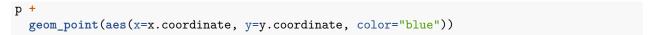
7.5.1 Exercises

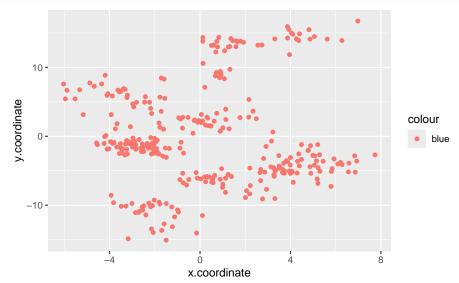
- 1. Try changing the cluster aesthetic to size and shape. Does this convey the same level of information as a color scale?
- 2. What other variable in our dataset would be well represented by a shape scale? Add a shape aesthetic for the variable you identified.
- 3. Generally speaking, which type of data lends itself to shape scales, which to size, which to color?
- 4. Why does this not work?

```
p +
    geom_point(aes(x=x.coordinate, y=y.coordinate, color=cluster)) +
    scale_color_brewer(type="qual", palette = "Set1")
#> Warning in RColorBrewer::brewer.pal(n, pal): n too large, allowed maximum for palette Set1 is 9
#> Returning the palette you asked for with that many colors
#> Warning: Removed 46 rows containing missing values or values outside the scale range
#> (`geom_point()`).
```



5. Why does this code not color all points in blue?





6. Advanced: Change the overall shape of points to number 24 triangles, than color by cluster and fill by type.

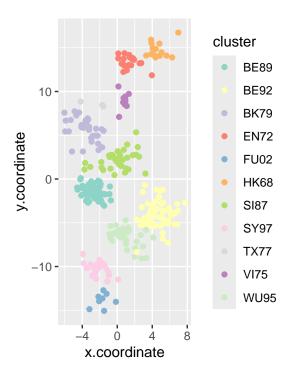
7.6 Setting labels and themes

So far, we have been concerned with the data layers of the plot, using <code>geom_xxx</code> to visualise different variables and <code>scale_xxx_yyy</code> to customise them. In the following section, we will have a look at customising the 'canvas' of the plot, i.e. the background, axis labels etc.

7.6.1 Coordinate system

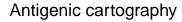
Looking at the antigenic map (Figure 1 in the Smith et al (2004) paper), we notice that their axis ratio of 1:1, ensuring that one unit on the y-axis is equivalent to one unit on the y-axis. Our plot has a different ratio. Based on the range of the x.coordinate and y.corrdinate variable, ggplot automatically chose the axis limits and more importantly here, their ratios. We can easily change this default by adding a coord_xx layer, in this case coord_fixed, which will ensure a 1:1 ratio per default.

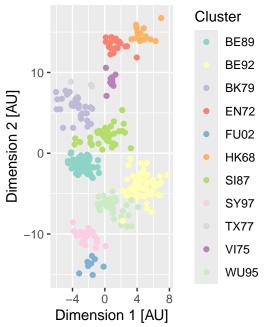
```
p +
    geom_point(aes(x=x.coordinate, y=y.coordinate, color=cluster)) +
    scale_color_brewer(type="qual", palette = "Set3") +
    coord_fixed()
```



7.6.2 Labels

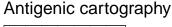
Currently, the axis and legend labels in our plot are simply the name of the variables we mapped to the aesthetics. We can change the axis labels by providing arguments to the labs layer. Specifically, x and y axis labels can be set by the xlab and ylab arguments respectively, the title is specified by the title argument. To change the name of a legend, we have to specify the aesthetic we mapped it to, in this color.

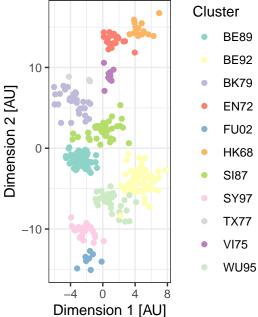




7.6.3 Themes

Finally, we can customise the non-data components of our plot. While the developers of ggplot2 had a strong preference for the default grey background, this is not to everyone's liking. As with everything else you've seen so far, there are some build-in options for customising. Here, we use the theme_bw to change to a white background.





7.6.4 Exercises

- 1. What other coordinate system options exist? Hint: type <code>?coord_</code> in a new chunk and press tab to see other options.
- 2. In the previous set of exercises, you added a shape aesthetic. Rename the legend title for this aesthetic
- 3. Test different themes and see how it effects the plot, for instance use theme_void, theme_dark and theme_classic. Similar to Exercise 1, you can type ?theme_ and tab to see other possible build in themes.
- 4. Why does this not work?

7.7 Saving your plots

You can save your plots with the ggsave function. ggsave will save the most recent plot to your project directory. As argument we only have to provide the name of the file we want to save it to. ggsave will determine the format of the output file based on the file ending of the filename that you provide. For instance, the code chunk below will save our most recent plot to a pdf document named "antigenic cartography.pdf".

```
ggsave(filename="results/antigenic_cartography.pdf")
#> Saving 6 x 3.71 in image
```

Not only does ggsave determine the format of the file from the name, it also determines the size of the file from the size we chose for displaying the plot in out analysis. To make it reproducible, it is good practice to specify the size and units.

Note: ggsave overwrites the previous file of that name without warning!

7.7.1 Exercises

- 1. Save the plot as png and jpeg.
- 2. Change the size of the plot with width and height; what happens to figure labels and legends?

8 Beyond scatter plots

In **section 4**, we have learned the basic recipes for generating a plot with **ggplot2**. We created a 2D scatter plot, encoding visual information in a color and shape scale and made sure we convey the right message by ensuring appropriate labels and coordinate systems.

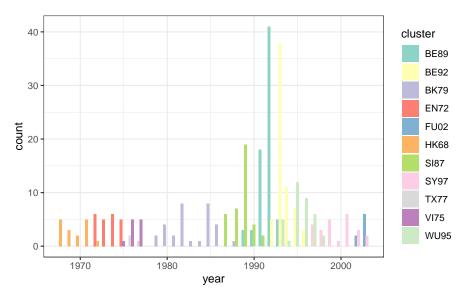
In the following section, we will get to know a couple more plot types.

8.1 Bar charts and histograms

Bar charts and histogram visualise the number of observations (count) for a specified variable, typically with the variable on the x-axis and the count on the y-axis. For histograms, the x-axis is divided into bins and the number of observations in each bin is counted. Bar charts are a special case of histogram, where the bin width is 1, i.e. the counts at each value of the variables are displayed. Bar charts are best described by the calling the help function <code>?geom_bar</code>:

```
There are two types of bar charts: geom_bar() and geom_col(). geom_bar() makes the height of the bar proportional to the number of cases in each group [..]. If you want the heights of the bars to represent values in the data, use geom_col() instead. geom_bar() [...] counts the number of cases at each x position. geom_col() [...] leaves the data as is.
```

In the following, we plot the number of antigen measurements per year and color them by cluster. As we want the plotting function to figure out the counts, we use <code>geom_bar</code>. For consistency to our previous plots, we use them same theme and color scheme; note, we use <code>geom_fill</code> (not <code>geom_color</code> as above) to fill the bars with the specified color. In addition to the aesthetics, we also specify positions for our bar chart. To display bars next to each other, we have to specify <code>position_dodge</code>, and setting <code>preserve="single"</code>, keeps a constant width for each bar. More on this in the exercises to this section.



We can use the same set-up with geom_histogram, to visualise the distribution of antigenic measurements in larger time intervals. The bin width specifies the range over which to summarise the variable. Again, the help function gives good insight with <code>?geom_histogram</code>:

binwidth [...] You should always override this value, exploring multiple widths to find the best to illustrate the stories in your data [...].

```
geom_histogram(aes(x=year, fill=cluster),
            position=position_dodge(preserve="single"),
            binwidth = 10) +
scale_fill_brewer(type="qual", palette = "Set3") +
theme bw()
                                                                            cluster
                                                                               BE89
               60
                                                                               BE92
                                                                               BK79
                                                                               EN72
            count
                                                                               FU02
                                                                               HK68
                                                                               SI87
                                                                               SY97
               20
                                                                               TX77
                                                                               VI75
                                                                               WU95
                        1970
                                     1980
                                                  1990
                                                              2000
                                           year
```

8.2 Boxplots

Below, we take a different look at the distribution of antigenic measurements by year, using a boxplot.

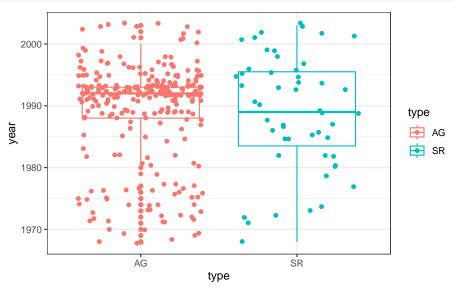
"The boxplot compactly displays the distribution of a continuous variable. It visualises five summary statistics

(the median, two hinges and two whiskers), and all "outlying" points individually." [?geom_boxplot].

We treat year as a continuous variable and show its distribution split by the type of measurement.

To show both the summary of the distribution (boxplot) and the actual data, we can add a geom_jitter to the plot. It plots the original y-values and adds a jitter to the x-values to avoid overplotting.

```
p + geom_boxplot(aes(x=type, y=year, color=type)) +
geom_jitter(aes(x=type, y=year, color=type)) +
theme_bw()
```



8.3 Geographical maps

So far, we have worked with visualisations of continuous, discrete and categorical variables. We have worked with the year (discrete/continuous), cluster, type (both categorical), x.coordinate and y.coordinate (both continuous) variables in out data set and used these to display the antigenic maps and distribution of measurements across time, separated by both cluster and type. For all these observations, we have an additional variable, the location of data generation. In the following, we will see how we can use ggplot2 to visualise geographic data.

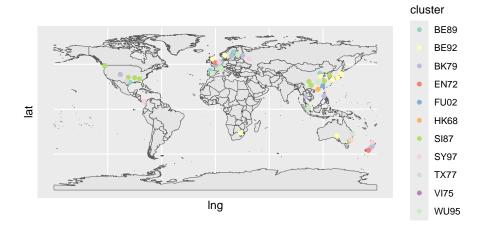
First, we read a file that contains the coordinates (latitude and longitude) of all locations that we find in the location of our coord object. We then use the ne_countries of the rnaturalearth package that we loaded in the beginning to create an object that countains coordinates of all countries.

As with above, we will then create a ggplot object. So far, we provided the data that we want the visualisation to be applied to in the ggpot call. In this case, we have two different data sets that we want to visualise, the world map and the locations. In this case, we can also specify the appropriate dataset to each geom separately.

We use the geom_sf to display the world map and a points layer with geom_point to visualise the locations.

```
world <- ne_countries(scale = "medium", returnclass = "sf")

g <- ggplot()
g + geom_sf(data = world) +
  geom_point(data=coord, aes(x=lng, y=lat, color=cluster)) +
  scale_color_brewer(type="qual", palette = "Set3")</pre>
```



8.4 Exercises

- 1. Test different options for the position argument of geom_bar. Hint: use the Details paragraph in ?geom bar to find a description about possible options.
- 2. What happens when you choose preserve="total" in position_dodge of geom_histogram?
- 3. Customise the color scale and plot labels in the boxplot showing the distribution of measurements per type and year. What happens if you choose aes(fill) instead of aes(color)?
- 4. Change geom_jitter to geom_point to see why geom_jitter is a better visualisation of the data. Go back to using geom_jitter and play with the width argument to customise your plot.
- 5. What would be a good theme for the world map? Add it to the plot.

9 Additional material

For future reading on data visualisation, selecting appropriates aesthetics and scales and avoiding common pitfalls, I recommend the book Fundamentals of Data Visualization Wilke (2019) (with free online version!).

A great overview of the most appropriate graph for your data type can be found on this website: From data to viz.

References

Smith, Derek J., Alan S. Lapedes, Jan C. de Jong, Theo M. Bestebroer, Guus F. Rimmelzwaan, Albert D. M. E. Osterhaus, and Ron A. M. Fouchier. 2004. "Mapping the Antigenic and Genetic Evolution of Influenza Virus." *Science* 305 (5682): 371–76. https://doi.org/10.1126/science.1097211.

Wilke, Claus O. 2019. Fundamentals of Data Visualization. 1st ed. O'Reilly Media, Inc. https://serialmentor.com/dataviz/.