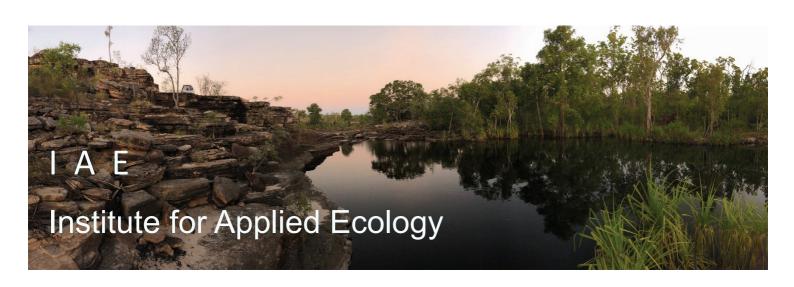


SNP Analysis using dartR

RStudio Refresher



Copies of the latest version of this tutorial are available from:

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RStudio Refresher

What is R?



R is a statistical computing language based on an earlier implementation of a programming language called S. S is still available in the commercial form of Splus, whereas R is in the public domain.

R was created by Ross Ihaka and Robert Gentleman (hence the name R) at the University of Auckland, New Zealand, and is now developed by the R Development Core Team.

Many statistical packages on the market, such as SAS, SPSS and Statistica are regarded as fourth generation statistical programming languages. The R programming language is a hybrid between a third-generation language such as C or FORTRAN and a fourth generation language such as SAS. This provides for much greater flexibility for the analyist, but demands much more in terms of programming skills.

R supports a wide variety of statistical and numerical techniques, with comparable benchmark results to Octave and its proprietary counterpart MATLAB. R is also provides the analyst with a very wide range of packages, which are user-submitted program **libraries**, for specific functions or specific areas of study. As a result, R is one of the most comprehensive statistical analysis systems on the market. R has exceptionally good graphical capacity, and can be used to produce publication-quality graphs.

The library we will be primarily using in this workshop is **dartR**, a collection of scripts to facilitate analysis of data provided by Diversity Arrays Technology Pty Ltd. Although dartR has some unique analyses, it is primarily for data manipulation, exploratory analysis, and a conduit to other packages used for SNP analysis.

The versatility of R has led to many different styles in the way the program is used. A programmer will use R in a very different way from someone using R to undertake statistical analyses. In this workshop, you will require familiarity with the R GUI, RSTudio, but will not necessarily need to be well versed in R programming.



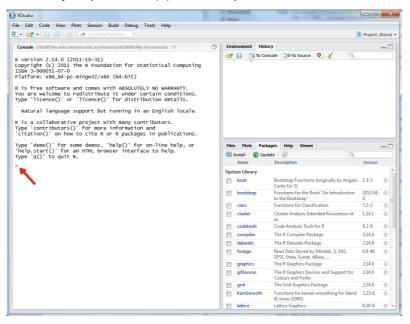


The Command Line Interface

Rather than using the standard distribution of R, we will be using an implementation with a graphical user interface called R-studio.

When you first start R-studio, a graphical user interface opens with many features to assist you (Figure 1-1). After some introductory text appears in the Console, a **Command Prompt** is presented (>), and the system awaits instructions.

Figure 1-1. R
as it appears
when it first
starts. The R
Console
window and
two other
windows are
visible. The
Program Editor
and R
Graphics
windows do
not appear
until required.



Before we move on, there are a couple of little tricks here that are worth mentioning. The first is that the Console can be cleared of text using control-L (^1). The second tip is that the up-arrow will recall previously submitted commands, which will save you a lot of typing. Try these as you go along.

The simplest way of using R is to supply instructions to this command prompt.

```
> sum < 125 + 172
```

Here we are adding two numbers and putting the answer in the scalar object (single valued vector) called sum.

You can view the contents of an object simply by giving its name in response to the command prompt.

```
> sum
[1] 297
```



Try some assignment statements for your self to undertake some arithmetic.

Here is a slightly more complex assignment statement.

```
> beetles <- c(15.2,12.1,17.8,13.9,16.4,15.1)
```

There is a lot to this simple command. What we are doing here is creating an ordered set of values, referred to as a **vector** in R terminology. In this case, the data are lengths of beetle elytra. The concatenate function c() is used to create the vector which is then assigned to the **object** beetles using the **assignment operator** <-. The object beetles is called an object because it is a self-contained entity that can be used in subsequent calculations.

Again, you can view the contents of an object simply by giving its name in response to the command prompt.

```
> beetles
[1] 15.2 12.1 17.8 13.9 16.4 15.1
```

Instructions to the command line are terminated with a return (¬) or a semi-colon (;). R instructions are case sensitive, so the objects beetles, Beetles and BEETLES are all considered as separate objects. It is wise to adopt a consistent practice, such as always using lower case unless upper case is demanded by the R syntax.

Spaces matter, sometimes. You will need to watch that.

If you instruct R to undertake some action, and do not assign it to an object, then R will direct the results of the instructions to the screen. For example, requesting R to create the vector of beetle elytra without assigning it to beetles will result in the vector being listed on the screen.

```
> c(15.2,12.1,17.8,13.9,16.4,15.1)
[1] 15.2 12.1 17.8 13.9 16.4 15.1
```

As an object, beetles can be used in subsequent calculations. For example,

```
mean (beetles)
[1] 15.08333
```



Try some assignment statements again, but this time directing the results to the screen. A bit like a simple calculator.

R programs often comprise a series of nested instructions, and the same result could have been obtained by using

```
> mean(c(15.2,12.1,17.8,13.9,16.4,15.1))
[1] 15.08333
```

This is the advantage of an object-oriented approach to programming.

The Graphics Interface

When a command requires more sophisticated output, R will open a purpose-built window. The most useful of these is the graphics window.

A scatter plot of 1000 pairs of coordinates drawn at random from a bivariate standard normal distribution (mean=0, stdev=1) is made by combining the plot() function with the rnorm() function as follows:

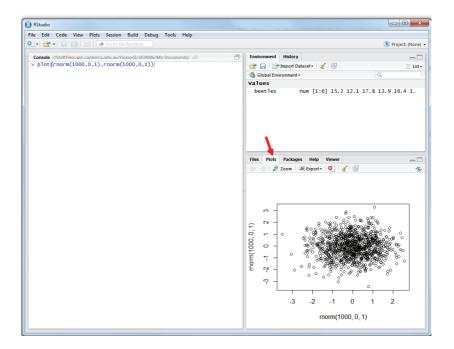
```
> plot(rnorm(1000,0,1),rnorm(1000,0,1))
```



Run this command to see if you can replicate the output below.

The result is shown in in Figure 1-2. This scatter plot can be saved to a file or copied to the clipboard by right-clicking on the graphics window and choosing the desired outcome.

Figure 1-2. R as it appears after activating the graphics window.



You can pull the graphics out into its own window with the [Zoom] tab, or export the image in one of the standard formats using the [Export] tab.

The R Editor Interface

Using the Command Line Interface is great for a quick analysis, but it is essentially a calculator mode. Once you have done the calculations, you walk away only with the results. In more substantial analyses, we need to better manage the set of programming instructions needed to do the job. We do this using the **R editor**, which can be accessed from the file menu

```
[File>New File>R Script]
or by typing
control-N (^N)
```



Open the RStudio Editor.

The idea is to type all instructions in the R editor for progressive submission or for submission as a block. At the end of the process, you have a complete program listing that can be saved to disk for later use.

The R editor is not all that sophisticated. Each instruction is typed in on its own line. A line can be submitted for execution by placing the cursor on it and typing control-enter (^_). Alternatively, blocks of instructions can be highlighted and submitted in the same way. This allows progressive debugging of the program as it is constructed.

It is wise to include abundant comments as part of your programs, so that you can understand them later or pass them to others in a comprehensible form. Comments are preceded by the # character and terminated by a return (\d).

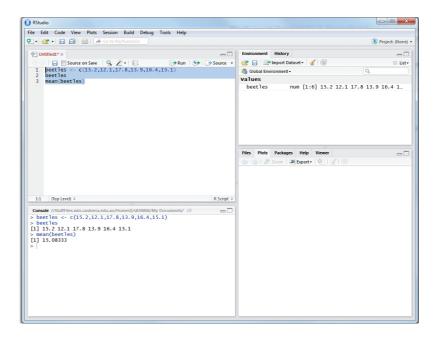
Our simple R program, with comments added is shown in Figure 1-3.

Creating and displaying a list of beetle measures
beetles <- c(15.2,12.1,17.8,13.9,16.4,15.1)
beetles
mean(beetles)</pre>



Type the above script into the RStudio Editor. Highlight the text and submit it for execution using control-Enter ($^{\sim}$ $_{\sim}$)

Figure 1-3. R as it appears after submitting a simple program.



You screen should look like that displayed in Figure 1-3.

Accessing Packages

Not many users of R program all the scripts that they require to undertake a task. This would be like reinventing the wheel. Instead, it is possible to access scripts written by those who have come before you, and who have made those scripts available as a package. A complete list of available packages can be obtained from the *Comprehensive R Archive Network* known as CRAN (https://cran.r-project.org/).

You will generally identify the packages you require after some investigative work on the web, by talking to colleagues or taking pointers from the literature. For example, the package reshape2 is useful for rearranging data, and can be accessed using the R-studio menus (Packages>Install Packages) or using the statement

> install.packages("reshape2")



Install reshape2

You may need administrator rights to install packages. Packages are only installed once, not every time you require them.

The directory where packages are stored is called the library. R comes with a standard set of packages. A list of installed packages can be obtained using

```
> library()
```

or by examining the list using the Packages menu.

Apart from those included in the standard implementation of R, installed packages are loaded for use in a session with the library function.

> library(reshape2)



Load the package reshape2

A list of loaded packages is obtained with

> search()

Error Handling and Help

When you make an error in the syntax of commands given to R, the program will respond with some form of diagnostic message. Sometimes these are self-explanatory, sometimes they are not.

Command line help

Fortunately, R has very extensive help documentation. If you know the exact name of the function you want help on (e.g. hist for plotting histograms), help can be obtained using

> ?mean

A window is displayed with help on the R function to generate the arithmetic mean. Note that the help gives you a list of possible parameters to pass to the function, and gives some simple examples of its operation.



Use the ? to generate help on the functions mean, sd, plot and matrix

Dynamic help

RStudio provides help on the fly. For example, as you are typing a function, the functions that begin with the letters you have typed are displayed as a menu. You can select the one you want, then hit tab to bring it in to your statement.



Help is also available by typing a tab after having selected a function. A list of parameters is displayed. Moving among the parameters gives help on each one.



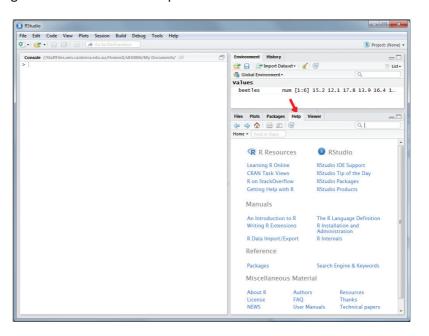


Try this for yourself

Help menu

More extensive help can be obtained from the help files using the [Help] tab of R-studio. Here you can use the Search Engine and Keywords link to access a wide range of information on the operations of R.

Figure 1-4. Useful information available using the [Help] tab.



Vignettes

Some packages in R have what are called **vignettes**. These are how-to guides for topics, and usually offer gentle introductions and examples. Alternatively, you can view vignettes from any loaded package by going to the 'Vignettes' menu and selecting the required package name. This will give a list of all available vignettes for you to open. Sometimes this menu doesn't appear until you load a package which has a vignette.

The Web

The web and Google are good places to turn for assistance. An excellent quick reference to R can be found on http://www.statmethods.net/

Managing Your Workspace



Starting a New Session

R facilitates the management of workflow by defining a **workspace** to hold your objects – vectors, dataframes, user-defined functions and the like. A workspace can be saved at the end of a session, and reloaded at a later time when you want to continue the analysis.

Managing workflow can be difficult in R, and we need some basic rules to minimize confusion.

- Identify discrete projects or analyses and create a separate Windows directory for each one. This way you will avoid having a jumble of objects from many analyses in your workspace.
- Tidy up after each session, by removing all unwanted and temporary objects, before saving your workspace.
- Use standard file naming conventions, such as filename.R for R programs, filename.csv for raw data files and filename.Rdata for R binary files.

Once you have started R, you need to start a new project using File>New Project. R will prompt you for a directory in which to save all temporary and working files, and the project image if you choose to save it later.

R may ask you to save existing work before opening a new project. You should do this if you have important work that has been executed in a previously open project.

Resuming a Session

To open an existing project use File>Open Project. This will prompt you for the working directory, scan it for a previously saved workspace, load that workspace if it exists and pass control back to you via the command prompt.

Either way, with an existing or a new project, you can now begin a new analysis, or continue an existing analysis confident that the objects, datafiles, and other associated elements of your analysis are a self-contained unit.

Terminating a Session

Exit a session by exiting from R, at which time you will be asked whether or not you wish to save your workspace.

Managing your Objects

The active objects associated with your workspace are listed when you select the [Global Environment] tab in R-studio.

In addition, there are a number of useful functions for managing your workspace.

- > setwd("c://R_analysis") Sets the default directory for files, and action that can also be done from the R-studio menus.
- > ls() provides a list of objects in your current workspace.

Setting a default directory

The best way to manage your work is to ensure that the files associated with each project is in a separate directory on disk. To set the default directory use

```
> setwd("C:/Users/username/Documents/R demo")
```

R will then look in the directory R_demo when locating a file to read, and to write a file. Note the direction of the backslashes in the file specification.

Set the working directory to somewhere useful, by adding the above line to your script in the RStudio Editor. Submit it for execution.

You can identify the location of the default directory, if you forget where it is, using

```
> getwd()
```

You can get a listing of the files in the working directory using

> dir()

Where have we come?



The above Session was designed to give you an overview of the operation of R through the RStudio graphical user interface. Having completed this Session, you should now be familiar the following concepts.

- R has available a Graphical User Interface (GUI) called R-studio, and within it, the R Console, R Editor Window, Graphics Output Window, and various Help Windows.
- R packages are first installed, then loaded to become available for use.
- R establishes a workspace. Managing the objects in that workspace is challenging for the new user of R, but proficiency will come with practice.
- R has abundant sources of help, including placing a ? in front of a command, using tab to list options for a function, referring to the vignette if one has been provided in the loaded packages, and of course, Dr Google.