MODULE 3

DATA PRODUCTION

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TATISTICAL INFERENCE IS THE PROCESS of making conclusions about a population from the results of a single sample. To make conclusions about the larger population, the sample must fairly represent the larger population. Thus, the proper collection (or production) of data is critical to statistics (and science in general). In this module, two ways of producing data – (1) Experiments and (2) Observational Studies – are described.

♦ Inferences cannot be made if data are not properly collected.

3.1 Experiments

An experiment deliberately imposes a *condition* on individuals to observe the effect on the **response variable**. In a properly designed experiment, all variables that are not of interest are held constant, whereas the variable(s) that is (are) of interest are changed among treatments. As long as the experiment is designed properly (see below), differences among treatments are either due to the variable(s) that were deliberately changed or randomness (chance). Methods to determine if differences were likely due to randomness are developed in later modules. Because we can determine if differences most likely occurred o randomness or changes in the variales, strong *cause-and-effect conclusions* can be made from data collected from carefully designed experiments.

3.1.1 Single-factor Experiments

A factor is a variable that is deliberately manipulated to determine its effect on the response variable. A factor is sometimes called an **explanatory variable** because we are attempting to determine how it affects (or "explains") the response variable. The simplest experiment is a single-factor experiment where the individuals are split into groups defined by the categories of a single factor.

For example, suppose that a researcher wants to examine the effect of temperature on the total number of bacterial cells after two weeks. They have inoculated 120 agars¹ with the bacteria and placed them in a chamber where all environmental conditions (e.g., temperature, humidity, light) are controlled exactly. The researchers will use only two temperatures in this simple experiment – 10°C and 15°C. All other variables are maintained at constant levels. Thus, temperature is the only factor in this simple experiment because it is the only variable manipulated to different values to determine its impact on the number of bacterial cells.

♦ In a single-factor experiment only one explanatory variable (i.e., factor) is allowed to vary; all other explanatory variables are held constant.

Levels are the number of categories of the factor variable. In this example, there are two levels -10° C and 15° C. Treatments are the number of unique conditions that individuals in the experiment are exposed to. In a single-factor experiment, the number of treatments is the same as the number of levels of the single factor. Thus, in this simple experiment, there are two treatments -10° C and 15° C. Treatments are discussed more thoroughly in the next section.

The **number of replicates** in an experiment is the number of individuals that will receive each treatment. In this example, a replicate is an inoculated agar. The number of replicates is the number of inoculated agars that will receive each of the two temperature treatments. The number of replicates is determined by dividing the total number of available individuals (120) by the number of treatments (2). Thus, in this example, the number of replicates is 60 inoculated agars.

The agars used in this experiment will be randomly allocated to the two temperature treatments. All other variables – humidity, light, etc. – are kept the same for each treatment. At the end of two weeks, the total number of bacterial cells on each agar (i.e., the response variable) will be recorded and compared between the agars kept at both temperatures.² Any difference in mean number of bacterial cells will be due to either different temperature treatments or randomness, because all other variables were the same between the two treatments.

⋄ Differences among treatments are either caused by randomness (chance) or the factor.

The single factor is not restricted to just two levels. For example, more than two temperatures, say 10° C, 12.5° C, 15° C, and 17.5° C, could have been tested. With this modification, there is still only one factor – temperature – but there are now four levels (and only four treatments).

3.1.2 Multi-factor Experiments – Design and Definitions

More than one factor can be tested in an experiment. In fact, it is more efficient to have a properly designed experiment where more than one factor is varied at a time than it is to use separate experiments in which

¹An agar, in this case, is a petri dish with a growth medium for the bacteria.

²Methods for making this comparison are in Module 17.

only one factor is varied in each. However, before showing this benefit, let's examine the definitions from the previous section in a multi-factor experiment.

Suppose that the previous experiment was modified to also examine the effect of relative humidity on the number of bacteria cells. This modified experiment has two factors – temperature (with two levels of 10° C or 15° C) and relative humidity (with four levels of 20%, 40%, 60%, and 80%). The number of treatments, or combinations of all factors, in this experiment is found by multiplying the levels of all factors (i.e., $2\times 4=8$ in this case). The number of replicates in this experiment is now 15 (i.e., total number of available agars divided by the number of treatments; 120/8).

♦ The number of treatments is determined for the overall experiment, whereas the number of levels is determined for each factor.

A drawing of the experimental design can be instructive (below). The drawing is a grid where the levels of one factor are the rows and the levels of the other factor are the columns. The number of rows and columns correspond to the levels of the two factors, respectively, whereas the number of cells in the grid is the number of treatments (numbered in this table to show eight treatments).

	Relative Humidity 20% 40% 60% 80			ty
	20%	40%	60%	80%
$10^{o}\mathrm{C}$	1	2	3	4
$15^{o}\mathrm{C}$	5	6	7	8

3.1.3 Multi-factor Experiments – Benefits

The analysis of a multi-factor experimental design is more involved than what will be shown in this course. However, multi-factor experiments have many benefits, which can be illustrated by comparing a multi-factor experiment to separate single-factor experiments. For example, in addition to the two factor experiment in the previous section, consider separate single-factor experiments to determine the effect of each factor separately (further assume that individuals (i.e., agars) can be used in only one of these separate experiments).

To conduct the two separate experiments, randomly split the 120 available agars into two equally-sized groups of 60. The first 60 will be split into two groups of 30 for the first experiment with two temperatures. The second 60 will be split into four groups of 15 for the second experiment with four relative humidities. These separate single-factor experiments are summarized in the following tables (where the numbers in the cells represent the number of replicates in each treatment).

Temperature			$R\epsilon$	elative	Humidi	ity
	$10^{o}\mathrm{C}$	$15^{o}\mathrm{C}$	20%	40%	60%	80%
ĺ	30	30	15	15	15	15

The tabel below was modified from the previous section to show the number of replicates in each treatment of the experiment where both factors were simultaneously manipulated.

	Relative Humidity			
	20%	40%	60%	80%
$10^{o}\mathrm{C}$	15	15	15	15
$15^{o}\mathrm{C}$	15	15	15	15

The key to examining the benefits of the multi-factor experiment is to determine the number of individuals that give "information" about (i.e., are exposed to) each factor. From the last table it is seen that all 120 individuals were exposed to one of the temperature levels with 60 individuals exposed to each level. In contrast, only 30 individuals were exposed to these levels in the single-factor experiment. In addition, all 120 individuals were exposed to one of the relative humidity levels with 30 individuals exposed to each level. Again, this is in contrast to the single-factor experiment where only 15 individuals were exposed to these levels. Thus, the first advantage of multi-factor experiments is that the available individuals are used more efficiently. In other words, more "information" (i.e., the responses of more individuals) is obtained from a multi-factor experiment than from combinations of single-factor experiments.³

A properly designed multi-factor experiment also allows researchers to determine if multiple factors interact to impact an individual's response. For example, consider the hypothetical results from this experiment in Figure 3.1-Left.⁴ The effect of relative humidity is to increase the growth rate for those individuals at 10°C (black line) but to decrease the growth rate for those individuals at 15°C (blue line). That is, the effect of relative humidity differs depending on the level of temperature. When the effect of one factor differs depending on the level of the other factor, then the two factors are said to *interact*. Interactions cannot be determined from the two single-factor experiments because the same individuals are not exposed to levels of the two factors at the same time.

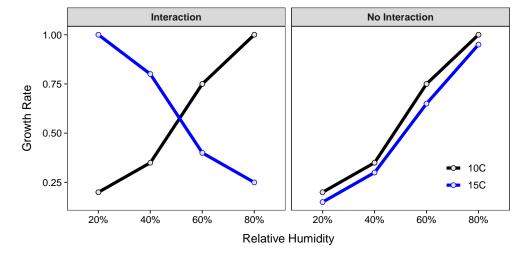


Figure 3.1. Mean growth rates in a two-factor experiment that depict an interaction effect (left) and no interaction effect (right).

Multi-factor experiments are used to detect the presence or absence of interaction, not just the presence of it. The hypothetical results in Figure 3.1-Right show that the growth rate increases with increasing relative humidity at about the same rate for both temperatures. Thus, because the effect of relative humidity is the same for each temperature (and vice versa), there does not appear to be an interaction between the two factors. Again, this could not be determined from the separate single-factor experiments.

³The real importance of this advantage will become apparent when statistical power is introduced in Module 12.

⁴The means of each treatment are plotted and connected with lines in this plot.

3.1.4 Allocating Individuals

Individuals⁵ should be randomly allocated (i.e., placed into) to treatments. Randomization will tend to even out differences among groups for variables not considered in the experiment. In other words, randomization should help assure that all groups are similar before the treatments are imposed. Thus, randomly allocating individuals to treatments removes any bias (foreseen or unforeseen) from entering the experiment.

In the single-factor experiment above – two treatments of temperature – there were 120 agars. To randomly allocate these individuals to the treatments, 60 pieces of paper marked with "10" and 60 marked with "15" could be placed into a hat. One piece of paper would be drawn for each agar and the agar would receive the temperature found on the piece of paper. Alternatively, each agar could be assigned a unique number between 1 and 120 and pieces of paper with these numbers could be placed into the hat. Agars corresponding to the first 60 numbers drawn from the hat could then be placed into the first treatment. Agars for the next (or remaining) 60 numbers would be placed in the second treatment. This process is essentially the same as randomly ordering 120 numbers.

A random order of numbers is obtained with R by including the count of numbers as the only argument to sample(). For example, randomly ordering 1 through 120 is accomplished with

```
> sample(120)
                                                                                                74
  [1]
        79
             78
                                          59
                                               35
                                                          8
                                                                                 38
                                                                                      81
                                                                                                     36
                  14
                       63
                            41 115 107
                                                  110
                                                              46
                                                                   64
                                                                        99
                                                                             37
                                                                                           48
             95
                  65
                       91
                                          97 108
                                                    62
                                                         39
                                                              42
                                                                   82
                                                                        47
                                                                                      29
                                                                                          113
                                                                                                 2
                                                                                                     53
 [21]
        77
                            26
                                98
                                       1
                                                                           101 106
 [41]
        18
             32
                  52
                       34 117 100
                                      43
                                          75 116
                                                    67
                                                         54
                                                              10
                                                                 102
                                                                        16
                                                                             92
                                                                                  88
                                                                                      40
                                                                                           17
                                                                                                     33
 [61]
        87
             70
                  13
                     111
                            89
                                85
                                      80
                                          83 112
                                                    86
                                                          6
                                                              19
                                                                   21
                                                                        84
                                                                             93
                                                                                 12
                                                                                      45
                                                                                           66
                                                                                                31
                                                                                                     30
 [81]
        22
             90
                  72
                        7 120
                                27
                                      50
                                          23
                                               71
                                                    69
                                                         25
                                                            103 109
                                                                        94
                                                                             15
                                                                                 55
                                                                                      58
                                                                                           61
                                                                                                60
                                                                                                     56
[101]
                       44 119 104
                                      68
                                          20
                                               51
                                                    24
                                                          5
                                                            114
                                                                        28 118 105
                                                                                                     49
                  73
```

Thus, the first five (of 60) agars in the 10°C treatment are 79, 78, 14, 63, and 41. The first five (of 60) agars in the 15°C treatment are 87, 70, 13, 111, and 89. In the modified experiment with two factors – temperature and relative humidity – with eight treatments containing 15 agars each, the random numbers would be divided into 8 groups each with 15 numbers.

```
> sample(120)
  [1]
        44
             34
                  90
                       27
                          100
                                      10 102 106
                                                      6
                                                             118
                                                                    67
                                                                         59
                                                                              80
                                                                                   42
                                                                                       97
                                                                                             46
                                                                                                 21
                                                                                                      41
 [21]
                  99
                            35
                                                     79
                                                                              78
                                                                                             13
                                                                                                      28
        53
             66
                      107
                                 25
                                      56
                                           29
                                                68
                                                          33
                                                               15
                                                                    62
                                                                        22
                                                                                   84
                                                                                         4
                                                                                                 12
 [41]
        65
             36
                 110
                       75
                            95
                                  1
                                      47
                                            8
                                                20
                                                     63
                                                          18
                                                             119
                                                                    11
                                                                        69
                                                                                    5
                                                                                       16
                                                                                            26
                                                                                                108
                                                                                                      49
                       19
                                                     32
 [61]
        39
             17
                  94
                            38
                                 14
                                      30
                                           60 117
                                                          54
                                                             114
                                                                    96
                                                                        50
                                                                              81
                                                                                  37
                                                                                       55 109
                                                                                                104
                                                                                                      82
 [81] 120
             43
                  72
                       57
                            93
                                 86
                                       2
                                           64
                                                40
                                                     45
                                                          23
                                                               24
                                                                    88
                                                                        91
                                                                               3
                                                                                  89
                                                                                       92
                                                                                           105
                                                                                                 48
                                                                                                      87
                                                                   71
                                                                                                      76
[101] 115
                  61
                                 77 112
                                           83 101
                                                     85
                                                          98 103
                                                                        73
                                                                               7 116
                                                                                        70
                                                                                             52
                                                                                                 58
             31
                       51 113
```

This design might be shown with the following table, where the numbers in each cell represent the first two agars selected to receive that treatment (only the first two are shown because of space constraints).

	Relative Humidity					
	20%	40%	60%	80%		
$10^{o}\mathrm{C}$	$44,\!34,\!\cdots$	$42,97,\cdots$	$33,\!15,\!\cdots$	$1,47,\cdots$		
$15^{o}\mathrm{C}$	$39,\!17,\!\cdots$	$37,55,\cdots$	$23,24,\cdots$	$77,112,\cdots$		

♦ Individuals should be randomly allocated to treatments to remove bias.

⁵When discussing experiments, an "individual" is often referred to as a "replicate" or an "experimental unit."

3.1.5 Design Principles

There are many other methods of designing experiments and allocating individuals that are beyond the scope of this book.⁶ However, all experimental designs contain the following three basic principles.

- 1. **Control** the effect of variables on the response variable by deliberately manipulating factors to certain levels and maintaining constancy among other variables.
- 2. Randomize the allocation of individuals to treatments to eliminate bias.
- Replicate individuals (use many individuals) in the experiment to reduce chance variation in the results.

Proper control in an experiment allows for strong cause-and-effect conclusions to be made (i.e., to state that an observed difference in the response variable was due to the levels of the factor or chance variation rather than some other foreseen or unforeseen variable). Randomly allocating individuals to treatments removes any bias that may be included in the experiment. For example, if we do not randomly allocate the agars to the treatments, then it is possible that a set of all "poor" agars may end up in one treatment. In this case, any observed differences in the response may not be due to the levels of the factor but to the prior quality of the agars. Replication means that there should be more than one or a few individuals in each treatment. This reduces the effect of each individual on the overall results. For example, if there was one agar in each treatment, then, even with random allocation, the effect of that treatment may be due to some inherent properties of that agar rather than the levels of the factors. Replication, along with randomization, helps assure that the groups of individuals in each treatment are as alike as possible at the start of the experiment.

3.2 Observational Studies – Sampling

In observational studies the researcher has no control over any of the variables observed for an individual. The researcher simply observes individuals, disturbing them as little as possible, trying to get a "picture" of the population. Observational studies cannot be used to make cause-and-effect statements because all variables that may impact the outcome may not have been measured or specifically controlled. Thus, any observed difference among groups may be caused by the variables measured, some other unmeasured variables, or chance (randomness).

Consider the following as an example of the problems that can occur when all variables are not measured. For many years scientists thought that the brains of females weighed less than the brains of males. They used this finding to support all kinds of ideas about sex-based differences in learning ability. However, these earlier researchers failed to measure body weight, which is strongly related to brain weight in both males and females. After controlling for the effect of differences in body weights, there was no difference in brain weights between the sexes. Thus, many sexist ideas persisted for years because cause-and-effect statements were inferred from data where all variables were not considered.

⋄ Strong cause-and-effect statements CANNOT be made from observational studies.

In observational studies, it is important to understand to which population inferences will refer.⁷ To make useful inferences from a sample, the sample must be an unbiased representation of the population. In other words, it must not systematically favor certain individuals or outcomes.

⁶Other common designs include blocked, Latin square, and nested designs.

⁷Thus, it is very important to first perform an IVPPS as discussed in Module 2.

For example, consider that you want to determine the mean length of all fish in a particular lake (e.g., Square Lake from Module 2). Using a net with large mesh, such that only large fish are caught, would produce a biased sample because interest is in all fish not just the large fish. Setting the nets near spawning beds (i.e., only adult fish) would also produce a biased sample. In both instances, a sample would be collected from a population other than the population of interest. Thus it is important to select a sample from the specified population.

⋄ It is important to understand the population before considering how to take a sample.

3.2.1 Types of Sampling Designs

Three common types of sampling designs – voluntary response, convenience, and probability-based samples – are considered in this section. Voluntary response and convenience samples tend to produce biased samples, whereas proper probability-based samples will produce an unbiased sample.

A **voluntary response** sample consists of individuals that have chosen themselves for the sample by responding to a general appeal. An example of a voluntary response sample would be the group of people that respond to a general appeal placed in the school newspaper. If the population of interest in this sample was all students at the school, then this type of general appeal would likely produce a biased sample of students that (i) read the school newspaper, (ii) feel strongly about the topic, or (iii) both.

A convenience sample consists of individuals who are easiest to reach for the researcher. An example of a convenience sample is when a researcher queries only those students in a particular class. This sample is "convenient" because the individuals are easy to gather. However, if the population of interest was all students at the school, then this type of sample would likely produce a biased sample of students that is likely of (i) one major or another, (ii) one or a few "years-in-school" (e.g., Freshman or Sophomores), or (iii) both.

In probability-based sampling, each individual of the population has a known chance of being selected for the sample. The simplest probability-based sample is the **Simple Random Sample** (SRS) where each individual has the same chance of being selected. Proper selection of an SRS requires each individual to be assigned a unique number. The SRS is then formed by choosing random numbers and collecting the individuals that correspond to those numbers.

For example, an auditor may need to select a sample of 30 financial transactions from all transactions of a particular bank during the previous month. Because each transaction is numbered, the auditor may know that there were 1112 transactions during the previous month (i.e., the population). The auditor would then number each transaction from 1 to 1112, randomly select 30 numbers (with no repeats) from between 1 and 1112, and then physically locate the 30 transactions that correspond to the 30 selected numbers. Those 30 transactions are the SRS.

Random numbers are selected in R by including the population size as the first and sample size as the second argument to sample(). For example, 30 numbers from between 1 and 1112 is selected with

```
> sample(1112,30)
 [1]
      106 1055
                 578
                       830 1066
                                  900
                                       525
                                             103
                                                  324
                                                        795 1057
                                                                   480
                                                                        773
                                                                              649
                                                                                    129
                                                                                         393
      215
           982
                                  200
[17]
                 628 1047
                            994
                                       863
                                             740
                                                  574
                                                             346 1003
                                                                              309
```

Thus, accounts 106, 1055, 578, 830, and 1066 would be the first five (of 30) selected.

There are other more complex types of probability-based samples that are beyond the scope of this course.⁸ However, the goal of these more complex types of samples is generally to impart more control into the sampling design.

♦ A proper SRS requires each individual i the population to be assigned a unique number.

If the population is such that a number cannot be assigned to each individual, then the researcher must try to use a method for which they feel each individual has an equal chance of being selected. Usually this means randomizing the technique rather than the individuals. In the fish example discussed on the previous page, the researcher may consider choosing random mesh sizes, random locations for placing the net, or random times for placing the net. Thus, in many real-life instances, the researcher simply tries to use a method that is likely to produce an SRS or something very close to it.

♦ If a number cannot be assigned to each individual in the population, then the researcher should randomize the "technique" to assure as close to a random sample as possible.

Polls, campaign or otherwise, are examples of observational studies that you are probably familiar with. The following are links where various aspects of polling are discussed.

- How Polls are Conducted by Frank Newport, Lydia Saad, and David Moore, The Gallup Organization.
- Why Do Campaign Polls Zigzag So Much? by G.S. Wasserman, Purdue U.

3.2.2 Of What Value are Observational Studies?

Properly designed experiments can lead to "cause-and-effect" statements, whereas observational studies (even properly designed) are unlikely to lead to such statements. Furthermore, in the last section, it was suggested that it is very difficult to take a proper probability-based sample because it is hard to assign a number to each individual in the population (precisely because entire populations are very difficult to "see"). So, do observational studies have any value? There are at least three reasons why observational studies are useful.

The scientific method begins with making an observation about a natural phenomenon. Observational studies may serve to provide such an observation. Alternatively, observational studies may be deployed after an observation has been made to see if that observation is "prevalent" and worthy of further investigation. Thus, observational studies may lead directly to hypotheses that form the basis of experiments.

Experiments are often conducted under very confined and controlled conditions so that the effect of one or more factors on the response variable can be identified. However, at the conclusion of an experiment it is often questioned whether a similar response would be observed "in nature" under much less controlled conditions. For example, one might determine that a certain fertilizer increases growth of a certain plant in the greenhouse, with consistent soil characteristics, temperatures, lighting, etc. However, it is a much different, and, perhaps, more interesting, question to determine if that fertilizer elicits the same response when applied to an actual field.

Finally, there are situations where conducting an experiment simply cannot be done, either for ethical, financial, size, or other constraints. For example, it is generally accepted that smoking causes cancer in humans even though an experiment where one group of people was forced to smoke while another was not allowed to smoke has not been conducted. Similarly, it is also very difficult to perform valid experiments

⁸For example, stratified samples, nested, and multistage samples.

on "ecosystems." In these situations, an observational study is simply the best study allowable. Cause-and-effect statements are arrived at in these situations because observational studies can be conducted with some, though not absolute, control and control can be imparted mathematically into some analyses. ⁹ In addition, a "preponderance of evidence" may be arrived at if enough observational studies point to the same conclusion.

 $^{^9\}mathrm{These}$ analyses are beyond the scope of this book, though.