CHAPTER 13 SCIENTIFIC INVESTIGATION

By the end of this chapter you will have covered the following material.

Science Inquiry Skills

- Identify, research and construct questions for investigation; propose hypotheses; and predict possible outcomes (ACSBL001 and ACSBL030)
- Desian investigations, including the procedure/s to be followed, the materials required, and the type and amount of primary and/or secondary data to be collected; conduct risk assessments; and consider research ethics, including animal ethics (ACSBL002 and ACSBL031)
- Conduct investigations, including using ecosystem surveying techniques, safely, confidently and methodically for the collection of valid and reliable data (ACSBL003)
- Conduct investigations, including microscopy techniques, real or virtual dissections and chemical analysis, safely, competently and methodically for the collection of valid and reliable data (ACSBL032)
- Represent data in meaningful and useful ways; organise and analyse data to identify trends, patterns and relationships; qualitatively describe sources of measurement error, and uncertainty and limitations in data; and select, synthesise and use evidence to make and justify conclusions (ACSBL004 and ACSBL033)

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- Interpret a range of scientific and media texts, and evaluate processes, claims and conclusions by considering the quality of available evidence; and use reasoning to construct scientific arguments (ACSBL005 and ACSBL034)
- Select, construct and use appropriate representations, including classification keys, food webs and biomass pyramids, to communicate conceptual understanding, solve problems and make predictions (ACSBL006)
- Select, construct and use appropriate representations, including diagrams of structures and processes; and images from different imaging techniques, to communicate conceptual understanding, solve problems and make predictions (ACSBL035)
- Communicate to specific audiences and for specific purposes using appropriate language, nomenclature, genres and modes, including scientific reports (ACSBL007 and ACSBL036)

Performing investigations is your chance to experience what doing science is really like. Science is about finding things out through observation and experiment, which is what doing investigations is all about. This is why investigations are central to science, *and* why they are so much fun.

Sometimes an important advance in science begins with a casual observation or a lucky accident. For example, after hearing from milkmaids that people who contracted cowpox (a relatively innocuous disease picked up after working with cattle) were protected from deadly smallpox, the British physician Edward Jenner effectively kick-started the science of vaccination. Jenner used samples from open cowpox sores on a dairymaid's hands to inoculate a young boy and protect him against smallpox. However, it would be another 50 years and a lot of carefully planned research before scientists truly began to understand the biological basis for immunity. This sort of lucky accident may begin a new field of research, but it then proceeds by carefully planned investigation.

Scientific investigations can take years to complete and may involve collaboration among many scientists. They may require access to special equipment in Australia or overseas. They may cost a lot of money, sometimes millions of dollars, to complete. Hence scientists invest time in *planning* investigations before they begin. When scientists apply for grants to carry out investigations they need to show that they have carefully planned what they will do and how any money provided will be spent. Good planning is crucial to the success of the investigation.

Scientists then make careful *measurements and observations* and record their *results*. They *keep records* of all their experiments. This is a legal requirement. Typically, experimental results need to be kept for 5–7 years. There are also requirements on how and where data is stored.

Once data is collected it needs to be *analysed*. There are various ways this is done, but in the biological and biomedical sciences it typically involves constructing graphs. Once a relationship is established graphically, a mathematical relationship can be derived.

Finally, the results of the investigation must be *communicated*. Usually this involves publishing a scientific paper either in a journal or conference proceedings. It often includes presenting the results in talks or posters at conferences. If the result is funded by a grant then a research report must be submitted. If the results are really exciting, then the scientists may write a media release. However the results are communicated, this step must happen for the investigation to be completed.

Planning your investigation

There are many things to consider when planning an investigation. You need to think about how much time you will have inside and outside class. You also need to think about the space and equipment you will need and where you will go if you want to make measurements or observations outside.

You may be working in a group or on your own. Most scientists work in groups. If you can choose who you work with, think about this carefully. It is not always best to work with friends. Think about working with people who have skills that are different from your own.

Finally, and probably the first thing that most students think about, is the topic of the research. You will need to come up with a **research question** or **hypothesis**.

Choosing a research question

Obviously, it is a good idea to investigate something that you find interesting. If you are working in a group try to find something that is interesting to everyone in the group.

A good way to start is by 'brainstorming' for ideas. This works whether you are working on your own or in a group. Write down as many ideas as you can think of. Don't be critical at this stage. Get everyone in the group to contribute and accept all contributions uncritically. Write down every idea.

After you have run out of ideas, it is time to start being critical. Decide which questions or ideas are the most interesting. Think about which of these it is actually possible to investigate given the time and equipment available. Make a shortlist, but keep the long list too for the moment. Once you have your shortlist it is time to start refining your ideas.

Researching and refining your question

The next step is to find out what is already known about the ideas on your list. Use the Internet, your text books and the library to find out. Make sure you *keep a record* of the information that you find as well as the *sources*. You should start a **logbook** at this stage. You can write in **references**, or attach printouts to your logbook. This can save you a lot of time later on! Many research students forget to do this when they first start reading about their topic and then have France of the search all over all of the search all over a search and equinoment. Once you have **Research in the search of the search students** forget to search all over again.

Good record keeping is important in scientific research, and it begins at this stage of the investigation.

Be critical of what you read. Do not assume that everything you read online or even in books is true. Try to find **reliable** sources of information. Textbooks, and websites from universities and government research agencies are usually very reliable. Publications and web pages from professional associations, such as the Commonwealth Scientific and Industrial Research Organisation (CSIRO), Australian Academy of Science and equivalent international organisations are also good sources. Blogs and homepages of other students are not usually reliable, although they are useful to give you ideas. Websites that are trying to sell you something should also be treated sceptically. Talk to your teacher about sources of information as well. They will be able to tell you if a website is reliable, and suggest sites that they know are suitable.

You may find examples of similar investigations to the one you are thinking of. It is a good idea to look at these, so you can learn from the experience other researchers. However, in general, it is better not to try to replicate someone else's investigation exactly. If you do decide to replicate someone else's investigation then you need to acknowledge and carefully reference their work. See the section on referencing below. If you do not do so, it is **plagiarism**. This is a very serious form of academic misconduct. Talk to your teacher about how original your research needs to be, and how closely it can be based on someone else's work. It is much better to do this at the start than to be accused of cheating later on!

Finally, talk to your teacher about your ideas. They will be able to tell you whether your ideas are likely to be possible given the equipment available. They may have had students with similar ideas in the past and can make suggestions.

After you have researched your questions and ideas, you will hopefully be able to narrow the shortlist down to the one question that you want to tackle. If none of the questions or ideas look possible (or still interesting), then you need to go back to the long list.

Figure 13.1 Brainstorm as many ideas as you can in your group.

CSIRO

This site contains useful information on all fields of scientific research, including resources for students and teachers.

AUSTRALIAN Academy of **SCIENCE**

This is a useful resource for up-todate science news.

Australian Institute of **BIOLOGY**

This is another useful resource that aims to promote education and research in Biology.

Proposing a research question or hypothesis

Once you have decided on what you will investigate, you need to turn it into a research question or a hypothesis. Make sure you check with your teacher what sort of investigation or project you are supposed to be doing.

A research question is one that can be answered by performing experiments or making observations. A hypothesis is a prediction of the results of an experiment, which can be tested by performing experiments or making observations.

You may also be able to do a 'design, build and test' project. These are described later.

Research questions

A research question may be of the form 'What effect does a new fertiliser have on root growth?' The aim of your research is to then answer the question. It is important that you frame the question carefully. It needs to be specific enough that it guides the design of the

investigation. A specific question rather than a vague one will make the design of your investigation much easier. Asking 'Does the new fertiliser increase root growth more than standard fertiliser?' tells you what you will be varying and what you will be measuring. It also gives a criterion for judging whether you have answered the question.

Asking 'How can we make roots grow the best?' is not a good question. This question does not say what will be varied, nor does it tell you when you have answered the question. 'Best' is a vague term. What you mean by 'best' may not be what someone else means.

A good research question identifies the **variables** that will be investigated. Usually you will have one **dependent variable** and one **independent** or **controlled variable**. For a lengthy investigation you may have two or more independent variables. Variables are discussed in more detail later.

Finally, a good research question should be answerable with the time and equipment available.

Hypotheses

A hypothesis is a tentative explanation or prediction, not yet confirmed by experiment, such as 'The new fertiliser makes roots grow longer in two weeks than standard fertiliser'. A hypothesis is often based on some existing **model** or **theory**. It is a prediction of what will happen in a specific situation based on that model.

A hypothesis should give you a prediction that you can test by performing an experiment. This means it should at least be **falsifiable**. A good hypothesis should be able to be disproved. However, you will *not* generally be able to claim that you have proved your hypothesis.

If your experiments agree with predictions based on your hypothesis, then you can claim that they support your hypothesis. This *increases your confidence* in your model, but it *does not prove that it is true*. Hence an aim for an experiment should not start 'To prove …', as it is not possible to actually prove a hypothesis, only to disprove it.

If your experimental results disagree, then you may have disproved your hypothesis. This is *not* a bad thing! Often the most interesting discoveries in science start when a hypothesis based on an existing model is disproved. This means that the model it was based upon is either not a good model, or does not apply to the particular situation. You could then try to work out why the model does not apply, or try to formulate a better model. What to do when your hypothesis is not supported is discussed further in the analysis section.

In summary, a good research question is a question that is specific and can be answered by performing experiments and making measurements. A good hypothesis is a statement that predicts the results of an experiment and can be tested using measurements.

Even if your question or hypothesis meets these criteria, do not be surprised if you change or modify it during the course of your investigation. In scientific research, the question you set out to answer is often only a starting point for more questions.

Designing your investigation

Once you have a specific research question or hypothesis, you need to design your investigation. It is fun to start making measurements or observations immediately, but it is also important to spend time learning how to use the equipment, and experimenting to find the best way to set up your investigation. You may also discover that you need different or more equipment. This may save you time later on.

It is also important not to get distracted and forget the purpose of your investigation. At the end of the process, you need good data that answers your question or tests your hypothesis. Having a plan ensures that you make the measurements that you need. The longer the investigation, the more important it is that you have a clear plan. There are several things to consider.

- What data will you need to collect?
- What materials and equipment will you need?
- When and where will you collect the data?
- If you are working in a group, who will collect the data?
- Who will be responsible for record keeping?
- How will the data be analysed?

The data that you collect will always include **secondary data**, and will usually include **primary data**. Secondary data is data that has been collected by someone else.

You will already have collected some secondary data when you investigated your research topic to formulate your question or hypothesis. You will probably want to collect more secondary data. If your topic is not one for which you can collect primary data, then you will need to rely on secondary data. Remember that when you collect secondary data it is important to use reliable, reputable sources.

Primary data is data that you collect yourself. You can collect data by performing experiments or making observations in the field. You should be able to measure parameters that are relevant to the biological question being asked (e.g. rate of cell division, body temperature, enzyme activity, size of population). You will have had practice at measuring some or all of these things already. You need to decide which variables you will measure and which variables you will control. Consider which variables you can control, and which you cannot.

Consider how you will analyse the data. Will you need access to specific software such as a graphing or statistics package? If so, make sure that you know how to use it. If you are using software to draw graphs then you need to know how to produce a **scatter graph** and fit a **line of best fit** and add **uncertainty bars**. Note that a line of best fit is *not* the same as joining the dots. You should *never join the dots*, even though this is often the default setting in spreadsheet software. You should consult a reference guide, the 'help' menu for your software, or ask your teacher. Graphs are discussed in more detail in the analysis section below.

Keep a record of your planning. This should go in your logbook. Writing down what you plan to do, and why, will help you stay focused during the investigation. If you are working in a group, then a record of what each person agrees to do during the investigation can be very important.

Variables and measurements

Anything that can vary in an experiment is a variable. An independent or controlled variable is one whose properties you can control. For example, if you were doing an experiment to measure the activity of an enzyme at different pH, then you would control the pH of the reaction and measure the rate of catalysis of the substrate. In this case the pH is the independent variable and the rate of catalysis, which varies with pH, is the dependent variable.

In the question 'Does the new fertiliser increase root growth more than standard fertiliser?', the type of fertiliser is the independent variable. The dependent variable is the root length (or change in root length). Other independent variables not mentioned in this question include soil composition, amount of water, light and temperature. These should all be kept constant, so they are not variables in the investigation. If it was a long investigation, the air pressure could be a second controlled variable. If you decide to have two independent variables then it is important to keep one constant while you vary the other, if at all possible. Then you take multiple sets of measurements, keeping one variable at a fixed value for each set of data while you vary the other.

When variables have a numerical value, you make **quantitative measurements**. You measure that numerical value in the appropriate units. For example, you may measure root length in centimetres or weight of roots in grams.

Continuous variables may take any possible value, usually within some range. Length, time and current are continuous. In the root growth example, root length is a continuous variable, as it will likely change over time. A variable that may take only fixed values is called a

Your measuring equipment will sometimes restrict you to only measuring discrete values. This is always the case with digital equipment. A set of digital scales that measures in grams gives you discrete values. It does not, however, mean that weight itself is a discrete variable. The weight of the roots is a continuous variable, but digital scales will only give you discrete measurements of the weight.

In some investigations you may use **qualitative measurements** or data. For example, a chemical reaction may lead to a colour change. You would usually describe the colour in words, such as 'pink' or 'green', rather than using a number. Sometimes you use a combination of qualitative and quantitative data. For example, you may describe the length of

Figure 13.3 ▼ Keeping growing conditions as consistent as possible helps to control the independent variables.

roots as reaching a maximum in centimetres (quantitative) but growing in a particular direction or pattern (qualitative).

Once you have decided on the variables you will be measuring, you will be able to identify the equipment and other resources you will need.

Identifying the resources required

If you are going to collect primary data, make a list of all the equipment that you need. Consider how precise the measurements will need to be. If your hypothesis predicts a temperature change of 0.1°C, but you can only measure to a precision of 0.5°C, then you will not be able to test your hypothesis. You may need to think carefully about how you measure some things. For example, in a root growth experiment, you may need to measure the dry weight of the roots, which means finding a consistent way to dry them.

Consider who will supply the materials and how much they might cost. Scientists generally have tight budgets that they have to work within. Also, the equipment you plan to use must be safe. Will you need special protective equipment, such as lab coats, safety glasses or gloves? There is a section on risk assessment below. Make sure that you include any safety equipment needed in your equipment list.

When you have your list, talk to your teacher about what equipment is available. You might find that you need to modify your question or hypothesis at this stage.

Consider where you will perform your experiments or observations. Can you use normal classroom space, or do you need to be outside? If you are outside, what provisions can be made for ensuring you can work without interference? Will you need to consider the convenience or safety of others? Talk to your teacher about what space is available.

Planning the experimental procedure

The most common problem that students have when doing research is time management. It is important to plan to have enough time to perform the experiments, *and* to analyse them, *and* to report on them. You also need to allow time to learn how to use the equipment if you have not used it before.

In any investigation you will need to collect reliable and precise data. You cannot do this if you do not know how to use the equipment. Always ask if you are unsure. Reading the user manual is also a good idea. It will usually specify the precision of the device, and let you know of any potential safety risks.

Whenever possible you should make repeat measurements, so allow time for this. This allows you to check that your measurements are **valid**. Valid results are affected only by a single independent variable. If the results are similar each time, then your results are likely to be valid. If a result is not **reproducible**, it is probably not a valid result. A result is reproducible if you make exactly the same measurement more than once and get the same result, within the limits of experimental uncertainty. If a result is not reproducible, then a variable other than the one you are controlling is affecting its value. If this is the case, you need to determine what this other variable is, and control it if possible.

Think about how you can minimise uncertainties. Minimising uncertainty is not just about using the most precise equipment you can find, it is also about clever experimental technique. Important discoveries are possible using simple equipment and techniques. For example, Gregor Mendel defined his laws of Mendelian inheritance through the careful, meticulous cultivation of pea plants.

Sometimes experiments simply don't work or can't be done for some reason such as equipment failure or unforseen variables. For example, root growth will be affected if the plants contract a disease during the experiment. Try to think of all the things that could go wrong. If possible, come up with backup plans. Allowing plenty of time helps with this, as does starting your experiments as soon as possible.

Make sure you allow time for analysis. Ideally, do as much analysis as you can while you collect results. If you plot graphs as you take measurements, then you will be able to identify **outliers** early. An outlier is a data point that does not fit the pattern of the rest of the data. If you identify an outlier while you still have access to equipment and space, you can check the

measurement and make sure that you didn't make a mistake or that the experiment hasn't been compromised by an uncontrolled variable.

After you have analysed your results, you need to write your report or communicate your findings in some other form. You need to plan ahead how this will be done. If you are working in a group, who will write which part of the report, and when? Who will proofread it? Who will be responsible for making sure all the parts fit together?

You may find a timeline useful. A timeline helps keep you on track, and reminds everyone of their responsibilities. If you are working in a group, get everyone to agree on it.

You can use the following table as a template.

Risk assessment

You may be required to complete a risk assessment before you begin your investigation. Even if this is not a requirement, it is a good idea to think about it. You need to think about three things.

- 1 *What are the possible risks* to you, to other people, to the environment or property?
- 2 *How likely is it* that there will be an injury or damage?
- 3 If there is an injury or damage to property or environment, *how serious are the consequences* likely to be?

A 'risk matrix', such as Table 13.1, can be used to assess the severity of a risk associated with an investigation. The consequences are listed across the top, from negligible to catastrophic. Negligible may be getting clothes dirty or a very minor injury such as a scratch. Marginal might be a bruise from falling off a bike, or a broken branch in a tree. Severe could be a more substantial injury or a broken window. Catastrophic would be a death or the release of a toxin into the environment. In general, you need to ensure that your investigation is low risk. You can use a risk matrix either for individual identified risks, or for the investigation overall. If there are multiple experiments, then you would use a risk matrix for each one.

$Consequences \rightarrow$ Likelihood	Negligible	Marginal	Severe	Catastrophic
Rare	Low risk	Low risk	Moderate risk	High risk
Unlikely	Low risk	Low risk	High risk	Extreme risk
Possible	Low risk	Moderate risk	Extreme risk	Extreme risk
Likely	Moderate risk	High risk	Extreme risk	Extreme risk
Certain	Moderate risk	High risk	Extreme risk	Extreme risk

Table 13.1 Matrix for assessing for severity of risk

Once you have considered what the possible risks are, you need to think about what you will do about them. What will you do to minimise them, and what will you do to deal with the consequences if something does happen? This may be as simple as 'Always wear a lab coat, gloves and safety glasses.' You can use a risk assessment table similar to the one shown.

Safe use and disposal of biological material

When dealing with many biological materials, it is important to be aware of safe handling and disposal. For example, when growing known or unknown microbes on agar plates, it is important to use safe sterile techniques (discussed below) and to wear lab coat, gloves, safety glasses and, if required, face masks. Treat all microbes on agar plates as potentially pathogenic and **autoclave** used plates before disposing.

Ethics

Ethics in research can be controversial. More than one scientist has lost their job for unethical research behaviour. Being ethical in your research has two aspects. The first is about being honest as a scientist. This means recording data accurately, and not ignoring, hiding or changing any data that doesn't support your hypothesis. It means acknowledging and referencing sources of information including books, websites, articles and people who have helped you. It means not using other people's ideas or data without their knowledge and permission. Put simply, it is showing integrity or 'doing the right thing'. A good rule is that if you wouldn't want someone to know what you are doing, you probably shouldn't be doing it. It is no different from behaving ethically in any other area of

your life. The other aspect to ethics is treating animals, other people and the environment with care and respect. If your investigation will be using humans then you need to make sure you do not harm them, either physically or psychologically. If you are working with animals, then you need to make a strong case for any investigation that harms or could potentially harm them. When scientists want to use humans or animals in their research, they need to be able to show that the benefits to the environment, other animals or humans significantly outweigh the negative effects on the animals or humans used.

The use and welfare of animals for the purposes of research is legislated by State and Federal laws and respect for all animals (vertebrate and invertebrate) used in research is of the utmost importance. When using animals for research, scientist must adhere to the '3Rs'. These are:

- replacement of animal research with other types of research where possible
- *reduction* of the number of animals used in research
- refinement of experimental techniques to minimise pain and distress. The National Institute of Health and Medical Research (NHMRC) has

guidelines on the ethical use of humans and animals in experimentation. If you are planning to collect live specimens in the field, be aware of

specific State laws that may pertain to native species (including plants).

Experimental technique

Once you have planned the experimental procedure, taking into account the minimisation of uncertainty, the risks involved and ethical considerations, it is time to begin the experiment.

In biology, a number of specialised techniques exist to ensure that the data collected is as reliable as possible. For example, in many laboratories, it is essential to use the **aseptic technique** to ensure that only the desired organisms grow in culture. Outside the lab, biological parameters can vary widely within populations and it is important to sample the population carefully to get an accurate representation of the population at large.

Aseptic technique

When dealing with cell culture (plant, animal and microbial), it is important to practice good aseptic (sterile) technique. This may mean working in a laminar flow hood – an enclosed workspace that prevents contamination of biological samples by maintaining positive pressure using filtered, sterile air. If a laminar flow hood is not available, a small sterile space can be

▼ Figure 13.4 The use of animals for research purposes is governed by State and Federal laws.

NHMRC human

and animal ethics guidelines here.

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created using a Bunsen burner, whose flame creates an updraft and kills airborne contaminants in the surrounding air.

Working in a sterile space must be combined with careful handling of all biological material and equipment and regular decontamination, typically with a solution of 80% ethanol.

Alternatively, when preparing microbial cultures on agar, utensils used to transfer the microbes may first be held over a flame to sterilise them, then cooled in a sterile environment before use. Holding the Petri dish upside down as much as possible minimises the opportunity for airborne contaminants to land on the agar.

Figure 13.5 **A** The use of a laminar flow hood can prevent contamination of biological samples.

Figure 13.6 **A** Tagging captured sea turtles, then performing recapture, helps estimate their population size from those sampled.

Population sampling

Taking accurate measurements in the field relies on measuring a large enough, representative sample of the population. Various standardised techniques have been developed to accurately sample populations and ecosystems. Different techniques are suitable for different types of studies and the method of sampling a population is an important component of the experimental design. For example, the **capture–mark–recapture** method is a powerful technique for estimating a population size, but relies on the captured sample redistributing equally throughout the population and being recaptured at the same frequency as individuals that had not been captured. It is important that such limitations be considered when planning your experiment.

Collecting your data

Once your experiment is underway, it is time to start collecting data. This is usually the fun part of any investigation. Don't forget you have a question to answer or a hypothesis to test! To do this, you need to make sure that you think carefully about what you do and keep good records.

Record keeping

You will need to keep a record of what you do during your investigation. You do this is in a logbook.

Scientists keep a logbook for each project that they work on. It is a record of what they did, why they did it, and what they found out. A logbook is a legal document for a working scientist. If someone's work is called into question, or there are disputes over patents or ownership of data, then the logbook acts as important evidence. Every entry in a scientists logbook is dated, records are kept in indelible form (pen, *not pencil*), and entries may even be signed. Scientists' logbooks include details of experiments such as methods and results. They include comments and ideas, thoughts about the experiments, and analysis. They frequently include printouts of data, photocopies of relevant information, photos and other items. The logbook is the primary source of information when a scientist writes up their work for publication.

Some scientists keep their research records electronically, but most experimental scientists still keep a hardcopy logbook. There are several advantages to a hardcopy logbook over an electronic one. First, electronic records are easy to make changes to, and it is hard to track what was changed, when and by whom. Second, if you are working in a group, it can be hard to keep track of who has the most recent version of the file/s. Third, files can be easily deleted or corrupted. It takes much more care and discipline to maintain a good electronic logbook than a good hardcopy. Remember that the purpose of a logbook is to record and maintain evidence of what you did. Electronic evidence is not as reliable as a signed hardcopy document.

You should talk to your teacher about what form of logbook records they require you to keep.

If you are working in a group then you will need to decide whether to keep one logbook for the entire group, or one each. If you will all be working in the same places at the same times, then one for the whole group is best. If you will be in different places (for example, doing field observations) then you will need one each. Your teacher may also require each of you to keep your own logbook for assessment, or for authentication purposes.

Your logbook is a detailed record of *what you did* and *what you found out* during your investigation. Make an entry in the logbook *every* time you work on your investigation. At the start of each session you should record the date and the names of all the people with whom you are working at the time.

Write down what you do as you do it. It is easy to forget what you did if you do not write it down immediately. An accurate record is important if you need to repeat any measurements or if you get unexpected results.

Include large, clear diagrams of any experimental set-up and include details of equipment used. You can also include photos of experiments.

Record the results of *all* measurements *immediately and directly into your logbook, in pen*. *Never* record data onto bits of scrap paper instead of your logbook! Results must be recorded in indelible form. This means using a pen. Never write your results in pencil. Never use white-out or scribble over anything in your logbook. If you want to cross something out, just put a line through it. It is also a good idea to make a note explaining why it was crossed out.

Performing experiments

If you have planned carefully and learned how to use the equipment, then hopefully your experiments will go smoothly.

The raw data should always be recorded directly into the logbook unless it is recorded using data loggers connected to a computer. In this case a printout of the data should be attached to the logbook, and the file name and location recorded. Make sure that you measure and record everything you will need for your analysis. For example, if you are investigating root growth, you could measure the amount of fertiliser, the temperature and the starting length of the roots. It is much better to measure something and then discover that you didn't need to, than to start your analysis and realise that you didn't measure something that you do need.

Use appropriate units; for example, centimetres for lengths and grams for weights. If you are going to be collecting multiple data points, it is a good idea to draw a table to record them in. Label the columns in the table with the name and units of the variables. Do not put the units in the table cells. Note that the accuracy of your measurements will often be restricted by the

instruments you use to take them. For example, a ruler may only have markings down to 0.1cm. Make a note of these restrictions as they may affect the accuracy of your final results, especially if the changes measured are very small.

If you have not made a mistake, then plotting and analysing as you go allows you to spot something interesting early on. You then have a choice between revising your hypothesis or question to follow this new discovery, or continuing with your plan. Many research projects start with one question and end up answering a completely different one. These are often the most fun, because they involve something new and exciting.

Analysing your data

When you have collected all your data you will need to analyse it. Record all your analyses in your logbook. If this is done on a computer, then record the file name and location and attach a printout of the analyses into your book. Many scientists have logbooks that are bulging with printouts.

The first step is organising your data. If you have more than a few data points, it is a good idea to display them in a table. You may have several tables for different experiments. You may also need to do some analysis of the data. For example, you may wish to show the change in root length over the course of the experiment in addition to the lengths at the beginning and end of the experiment.

Plotting graphs is a useful way to begin the analysis of your data. Graphs are a very useful way of representing data so that trends and relationships can be identified. There are many different sorts of graphs that can be used to organise and display data. These are described below. You will usually need to do some calculations with your data to be able to answer your question or test your hypothesis. Remember to keep units on all quantities, so that any derived values have the correct units. You will also need to calculate uncertainties on any derived quantities.

Identifying trends, patterns and relationships

You may be able to see a pattern simply by looking at a list of numbers in a table. However, the most reliable way to identify a pattern in data or a relationship between variables is to plot a graph.

A graph should be large and clear. The axes should be labelled with the names of the variables and their units. Choose a scale so that your data takes up most of the plot area. This will often mean that the origin is not shown in your graph. Usually there is no reason that it should be.

When you are looking for a relationship between variables, plot a scatter graph. This is a graph showing your data as points. Do not join them up as in a dot-to-dot picture. Usually the independent variable is plotted on the *x*-axis and the dependent variable goes on the *y*-axis, unless there is a good reason to do otherwise. For example, if you were measuring root growth in response to temperature, root growth (change in length) would be plotted on the *y*-axis against temperature on the *x*-axis.

To determine a relationship you need to have enough data points and the range of your data points should be as large as possible. A minimum of six data points is generally considered adequate if the relationship is expected to be linear, but always collect as many as you reasonably can, given the available time. For non-linear relationships you need more data points than this, so collect as many as possible.

Figure 13.7 **▲**

Plan exactly what you will measure to collect your data. Do you want to test how to grow longer roots, or a greater mass of roots? Where do the roots end? Will you use fresh weight or dry weight?

Data points

This website contains some helpful advice on deciding the number of data points.

A good graph to start with is simply a graph of the raw data. You will usually be able to tell by looking whether the graph is linear. If it is, then fit a straight line using a graphing package. You can then use a **linear regression** tool to check how good the straight line fit is. This will give you an R^2 number, which is a measure of 'goodness of fit'. The closer R^2 is to 1 (or –1), the better the fit. If it is not *very* close to 1, then the relationship is not linear.

If it is a linear relationship, then finding the equation for the line of best fit may be useful. *Never* force a line of best fit through the origin. Often the intercept gives you useful information. It may even indicate a systematic error, such as a zero error in calibration of your equipment.

When you plot your raw data you may find that one or two points are outliers. These are points that do not fit the pattern of the rest of the data. These points may be mistakes; for example, they may have been incorrectly recorded or a mistake was made during measurement. They may also be telling you something important. For example, if they occur at extreme values of the independent variable then it might be that the behaviour of the system is linear in a certain range only. This is the case for many biological **assays**. You may choose to ignore outliers when fitting a line to your data, but you should be able to justify why.

When you extend a line of best fit beyond your measured points, this is called **extrapolation**. Any data that you read off a graph outside the range of your data points is extrapolated, and should be viewed with caution. You cannot say for sure that the system continues to behave in the same way beyond the bounds of your data.

Reading points, other than data points, from a line of best fit within the region in which you have data is called **interpolation**. You cannot be sure that this is exactly what you would find if you measured that point. However, if your line of best fit really represents the behaviour of the system, then you can use interpolated points in your analysis.

For example, an assay that measures the concentration of protein in a solution involves creation of a standard curve from which the concentration of samples can be interpolated (Figure 13.8). Since the assay has a minimum level of detection (its **sensitivity limit**) and a maximum detection limit above which the assay is saturated, it is not possible to extrapolate outside the linear region of the standard curve. To measure samples whose concentration is higher than the detection limits for the assay, the samples must be diluted so their concentration lies within the range of the assay (the linear region of the standard curve).

In biological experiments, you are often comparing the effects of different independent variables on a single dependent variable. In the root growth example, you are comparing the effect of the new fertiliser with those of the standard fertiliser. In this case, you would typically plot a bar graph of the change in root length against the type of fertiliser used. If the data is taken from several different plants in each treatment group, you would plot the mean plus or minus one **standard deviation** (Figure 13.9). You might then use appropriate statistical analysis to determine whether or not there is a **significant difference** between the treatments.

▲ Figure 13.8

Standard curves can be used to interpolate and extrapolate values, but only within the limits of detection.

Bar graph of two data sets, with standard

deviation

For data of this type, you might, for example, use a **t-test** to test for a significant difference between the two treatments. Graphing software typically has functions for calculating standard deviation and for statistical analysis of the data.

Interpreting your results

Once you have analysed your results you need to interpret them. This means being able to either answer your research question or state whether your results support your hypothesis. If you have performed statistical analysis, does this support your hypothesis? For example, if the new fertiliser induces statistically greater root growth than the standard fertiliser, with all other variables being equal, this

would support the hypothesis that 'The new fertiliser makes roots grow longer in two weeks than standard fertiliser'. If there was no difference between the two, or if the new fertiliser induced significantly less root growth than the standard fertiliser, then this would argue against the hypothesis.

If your hypothesis is not supported

It is not enough to simply say 'our hypothesis is wrong'. If the hypothesis is wrong, *what* is wrong with it?

It may be that you have used a model that is too simple, or did not take into account all of the other variables. For example, in the root growth experiments, it may be that the new fertiliser works best at a particular temperature, or over a longer time, or in conjunction with certain soil conditions. Or maybe it doesn't work with the type of plant you chose to use. It may be that the experiment was simply too limited to fully test the hypothesis. Thus, you might conclude that further experiments are required to test these other variables.

Before you decide that the model is at fault, however, it is a good idea to check carefully that you have not made any mistakes or ignored any variables.

Think carefully about any factors that you did not take into account but which might have affected your experiment.

Go through your method, results and analysis. Check that your equipment was correctly calibrated, and that you were using it correctly. Check that data is recorded in the correct units, and that units are correctly carried through all calculations during analysis. Check your analysis carefully. If you are working in a group, get another person to repeat the calculations.

It is never good enough to conclude that 'the experiment didn't work'. Either a mistake was made or the model used was not appropriate for the situation. It is your job to work out which.

Communicating your results

If research is not reported on, then no-one else can learn from it. An investigation is not complete until the results have been communicated. Most commonly, a report is written.

Writing reports

A report is a formal and carefully structured account of your research. It is based on the data and analysis in your logbook. However the report is a *summary*. It contains only a small fraction of what appears in the logbook. Your logbook contains all your ideas, rough working and raw data. The report typically contains none of this.

A report consists of several distinct sections, each with a particular purpose. These are:

Abstract Introduction Method Results and analysis Discussion Conclusion Acknowledgements References Appendices

Reports are always written in the past tense, because they describe what you have done.

The abstract

The abstract is a very short summary of the entire report. It is the most important part, because often it is the only part that people read. Typically an abstract is between 50 and 200 words long. It appears at the start of the report, but is always the last thing that you write. Try writing just one sentence to summarises each part of your report.

Introduction

The introduction tells the reader why you did the investigation and what your research question or hypothesis is. This is the place to explain why this research is interesting or important.

The introduction also provides any background information needed to be able to understand the rest of the report. This is the place to summarise any existing theories and models. You need to do this to justify your hypothesis. You should also summarise any similar investigations. All of this should be correctly referenced, as described in the section on referencing below.

Method

The method describes what you did. It is not a recipe for someone else to follow.

The method summarises what you measured and how you measured it. It also explains, briefly, why you chose a particular method or technique.

Write your method using sentences, not dot points. Remember that these need to be written in past tense – *it is not a recipe*. You are not commanding anyone to do anything. You are telling people what you did. For example, you would write 'we measured the length' not 'measure the length'.

Include any diagrams, such as circuit diagrams, that are needed to make your method clear. The diagrams in your logbook will usually be rough sketches. The diagrams in your report should be very neat and carefully labelled. Flow charts can be useful to describe any procedures in which a series of steps was followed. Each diagram should have a figure number and you should refer to it in the text of your report. Position the diagram close to where it is referred to in the text. You should take the time to learn how to position figures neatly using your word processor software. When including images taken on a **microscope**, a scale bar and magnification must always be noted.

Results and analysis

The results section is a *summary* of your results. It is usually combined with the analysis section, although they may be kept separate.

Avoid including tables of raw data in your report unless they compare the results of a few different experiments. Wherever possible use a graph instead of a table.

If a table has more than a few rows of data, it is better to represent that data in some other way. Usually this will be a graph.

Think about what sort of graph is appropriate. If you want to show a relationship between two variables then use a scatter plot. Display your data as points with uncertainty bars and clearly label any lines you have fitted to the data. Always make sure you label your axes, including units. Choose an appropriate scale so that the data takes up most of the plot area.

Column and bar charts are useful for comparing two data sets, such as average root length with different types of fertiliser. *Do not* use a column or bar chart to try to show a mathematical relationship between variables.

Figure 13.10 gives examples of the two types of graphs.

Figure 13.10 ▲

a) A scatter plot with linear regression demonstrating a mathematical relationship; b) A column graph comparing numbers between different groups

Any data and derived results should be given in appropriate units with standard errors of the mean or standard deviation, as appropriate. If you performed calculations or statistical analyses of the data, show the equations or describe the statistics you used. You might want to show one example calculation, but do not show more than one if the procedure used is repeated.

Discussion

The discussion should explain *what your results mean*. If you began with a research question, give the answer to the question here. If you began with a hypothesis, state whether or not your results support your hypothesis. If not, explain why. (You might only be able to say that the model was not suitable for the situation being investigated.)

If there are any implications of your work, such as implications for better agricultural processes or the design of better medicines, put them here.

The discussion is also the place to briefly describe any difficulties that you had and make suggestions for improving the process. Remember that you should never say 'the experiment didn't work' if you didn't get the results you expected. You might choose to make some comments on possible further work that could be done.

Conclusion

The conclusion is a *very* brief summary of the results and their implications. Say what you found out and what it means. A conclusion should only be a few sentences long. It should clearly refer back to your hypothesis or research question.

Acknowledgements

You should thank anyone who helped you in your investigation. This includes people who supplied equipment or funding, as well as people who gave you good ideas or helped with the analysis. In science, as in other aspects of your life, it is polite to say thank you; however, this is not a compulsory section of a report.

References

A reference list details the sources of all information that were actually used to write the report. Wherever a piece of information or quotation is used in your report it must be referenced *at that point*. This is typically done either by placing a number in brackets at the point [2], or the author and year of publication (Smith, 2014). The reference list is then provided either in a footnote at the end of the page or a single, complete list at the end of the report. Referencing must be done in a consistent style. Check with your teacher what style they prefer. There are several good online guides to referencing.

A reference list is *not* the same as a bibliography. A bibliography is a list of sources that are useful to understanding the research. They may or may not have actually been used by the report authors. You should have a bibliography in your logbook from the planning stage of your investigation. The references will be a subset of these sources.

Appendices

Appendices may be used to provide additional information, such as raw data that is not necessary to understanding the report but which might be of interest to some readers. Your teacher might require you to provide raw data in an appendix. Reports do not always have appendices.

Other ways of communicating your results

You may want to present the results of your investigation in some other way. Scientists communicate their work in many ways. Sometimes a poster is presented or a seminar is given. An article may be written or a website produced. Scientists usually use more than one means, and sometimes several of them, to communicate about a really interesting investigation.

REFERENCING

GUIDE

This guide is designed to help you with referencing your sources for assignments.

REFERENCING i-TUTORIAL

This tutorial will help you understand referencing and show you how to avoid plagiarism.

Report **WRITING** example 1

This online resource guides you through the sections of a typical report.

Figure 13.11

A poster session is a common way to present scientific findings at a conference.

Report

WRITING example 2 This online

resource will help you write a case study.

WEBSITE ACCESSIBILITY

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The Royal Society for the Blind has information on making websites accessible.

Look at examples of articles in the scientific and the popular media, on websites, posters and so on. This will give you an idea of the different styles used in the different modes. Think about the purpose. Is it to inform, to persuade or both? What sort of language is used?

Think about your audience and use appropriate language and style. A poster is not usually as formal as a report. A website may be more or less formal, depending on your audience.

Posters and websites use a lot of images. Images are usually more appealing than words and numbers, but they need to be relevant. Make sure they communicate the information you want them to.

Make sure you keep readability and accessibility in mind if you are creating a poster or website. Posters should use large clear fonts and not have too much text. They should be readable from a few metres away. Fonts also need to be large enough and clear on websites, and digital images should have tags. Refer to the *Website accessibility* weblink for more information on accessibility and web-page design.

However you communicate your work, make sure you know what the message is and who the audience is. Once you have established that, you will be able to let other people know about the interesting things you have discovered in your investigation.

CHAPTER GLOSSARY

aseptic technique the technique of working under sterile conditions to prevent contamination of samples

assay an experimental technique or procedure used to test a specific biological process or effect; for example, an enzyme kinetic assay

autoclave a device used to sterilise equipment, reagents or contaminated waste; autoclaves work by subjecting contents to pressurised steam at 121°C for a set time

capture-mark-recapture an ecological surveying technique used to measure animal populations, in which individual animals are captured, marked and released; after a time, the population is re-sampled and the number of marked animals caught gives an indication of population size

continuous variable a variable that is able to take any value within a range; length, time and temperature are examples of continuous variables

controlled variable the variable that is controlled by the experimenter, so that its values are chosen; also called the independent variable

dependent variable the variable that changes as a result of changes to the independent or controlled variable

discrete variable a variable that may take only certain values; number of individuals, or number of legs on an animal are examples of discrete variables

extrapolation extension beyond the measured range of data to read or construct new data that has not been measured

falsifiable able to be disproved

hypothesis a tentative prediction, usually based on an existing model or theory; also a tentative explanation of an observation based on an existing model or theory

independent variable a variable upon which another variable depends; usually the controlled variable

interpolation to read or construct a new data point that has not been measured but is within the range of measured data

linear regression a statistical tool used to model the dependence of one variable on another

line of best fit the line that most accurately fits the data, usually calculated using linear regression

logbook the record of an experiment or investigation kept by the scientist performing the experiment; it is a legal record of the experiments and their results

model the artificial conceptual or abstract simulation of a real-world process or system, developed by simplifying key steps that produce reliable and consistent agreement as verified by field studies; a model may be mathematical equations, a computer simulation, a physical object, words or other form

outlier a data point that does not fit the pattern shown by other measured data points

plagiarism presenting someone else's work, including their words or ideas, as your own

primary data data that you have measured or collected yourself

qualitative measurement a measurement with descriptive or non-numerical results

auantitative measurement a measurement with numerical values

reference the source of a specific piece of information or quotation

reliable highly likely to be true; a trustworthy source of information or reproducible data

reproducible giving the same result, within uncertainty limits, when repeated measurements are made

research question the specific question that a particular experiment or investigation is attempting to answer

scatter graph a graph or plot showing data points, without a line joining the points, and used to demonstrate or determine a mathematic relationship between variables; the axes are defined by the variables

secondary data data or information that has been collected by someone else

sensitivity limit the portion of a curve that is nonlinear; data that falls into these non-linear regions cannot be extrapolated

significant difference a difference between data values that is statistically significant; that is, the probability (*p*) of the difference being due to chance is so small (usually less than 5%) that the result is considered true

standard deviation a measure of the dispersion of a set of data from its mean; expresses the variability of a population or set of data

t-test a statistical test commonly used to analyse differences between two sets of data

theory a collection of models and concepts that explain specific systems or phenomena; scientific theories allow predictions to be made and hence are falsifiable

uncertainty bars bars drawn above and below and/or to left and right of a data point on a graph to indicate the size of the uncertainty in that point

valid results that are affected by only a single independent variable and hence are reproducible

variable something that can change or be changed, as distinct from a constant, which does not change