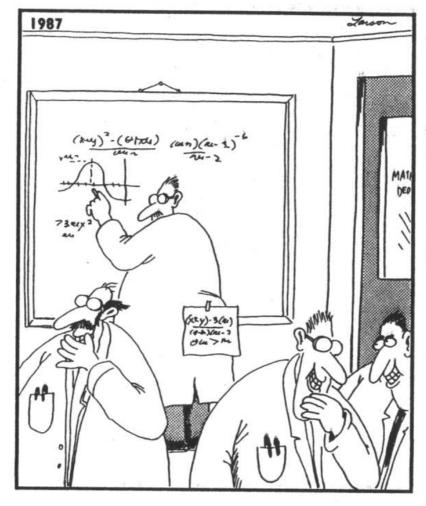
Merchant Taylors' School



Bíology A-Level

A2 Core Practical Workbook

Edexcel Specification



- 5.11 How to study the ecology of an area (see coursework)
- 5.17 How temperature affects the development of organisms
- 6.6 Polymerase Chain Reaction (PCR)
- 6.7 Electrophoresis
- 6.18 Which antibiotic is the most effective?
- 7.6 Investigating the rate of respiration
- 7.14 Spirometer and exercise
- 8.15 Habituation to a stimulus

<u>Some key expressions:</u>

Control Variable:	A factor that is kept constant so that its effects on the dependent variable are consistent throughout all experiments
Independent Variable	: The factor that affects the dependent variable. The factor you change.
Dependent Variable:	The factor that is affected by the independent variable. The factor you measure.
Reliability:	The same results are recorded if the experiment is repeated. Standard deviation and / or standard error are an excellent measures of reliability.
Accuracy:	There is little difference between your results and the recorded "true" results

- Validity: A combination of accuracy and reliability. Valid results are representative and can be used to make accurate predictions.
- Random Error: A mistake in the method or malfunction in the equipment which leads to the production of a single anomalous result, inconsistent with the trend. Once spotted, a random error should be either repeated or ignored.
- Systematic Error: Usually down to an uncontrolled factor, a affects the entire systematic error experiment, usually shifting the results by a experiment. consistent amount each Systematic always produce errors inaccurate results, but in some cases the data produced may still be reliable; and, as a trend may still be observable, valid to a degree.
- Null Hypothesis: The opposite of your working hypothesis: i.e. that the independent variable has no effect on the dependent variable. You aim to disprove this hypothesis in your experiment.
- **Experimental Hypothesis**: Your working hypothesis that the independent variable does have an effect on the dependent variable. By disproving the Null Hypothesis you can accept your Experimental Hypothesis¹.

¹ Note: it is virtually impossible to prove something correct, yet very simple to prove something incorrect. Therefore, scientists aim to disprove their Null Hypothesis, which then allows them to accept their Experimental Hypothesis according to the principle of Occam's Razor.

Part of Biology A-level is an assessment of "How science works"- an overview of the scientific process, neatly summarized into 12 criteria. You can be asked questions relating to these criteria in any written paper, so look at them carefully!

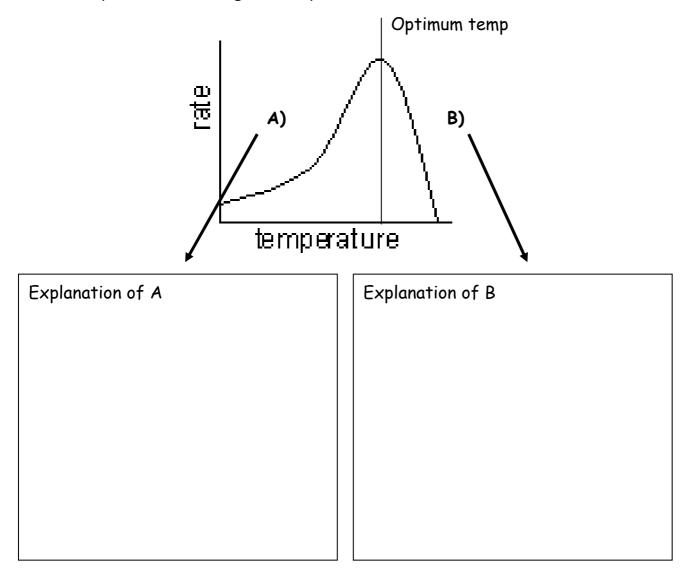
Each Core Practical has been designed to introduce you to some of these criteria. As you complete the practicals you should make a note in the table of which criteria the practical meets.

Criteria	Learning Outcome	Practical
1) Use theories, models and ideas to develop and modify scientific explanations	 a) Explain how the development of scientific theories involves hypothesizing, collecting and interpreting data and using creative thinking b) Explain the importance of modeling as a way of developing scientific understanding 	
2) Use knowledge and understanding to pose scientific questions, define scientific problems, present scientific arguments and scientific ideas	 a) Distinguish between questions that science can address, and those which science cannot address b) Identify scientific questions or problems within a given context c) Apply scientific theories to answer scientific questions or address scientific problems 	
3) Use appropriate methodology, including ICT, to answer scientific questions and solve scientific problems	Justify methods, techniques and processes used during scientific investigations, including use of ICT, to collect valid and reliable data and produce scientific theories for a chosen question or problem	

Criteria	Learning Outcome	Practical
4) Carry out experimental and investigative activities, including appropriate risk management, in a range of contexts	Produce a risk assessment before carrying out a range of practical work	
5) Analyse and interpret data to provide evidence, recognizing correlations and causal relationships	a) Analyze data including use of; - Descriptive statistics (mean, mode and median, error bars, SD, identification of outliers and range)	
	- Graphic representation to identify patterns and relationships (e.g. correlation and cause)	
	b) Interpret data with reference to the methods of the analysis used	
6) Evaluate methodology, evidence and data, and resolve conflicting evidence	Evaluate the validity of inferences made from data in terms of the methods, techniques and processes used to collect and analyze the data, recognizing any systematic or random errors present or conflicting evidence	
7) Appreciate the tentative nature of scientific knowledge	Explain how scientific theories are developed, refined, supported or refuted as new data or new interpretations of data become available	
8) Communicate information and ideas in appropriate ways using appropriate terminology	Present scientific information using text, graphics and other media as appropriate using scientific terminology with reference to data and credible sources	

Criteria	Learning Outcome	Practical
9) Consider applications and implications of science and appreciate their associated benefits and risks	a) Evaluate activities in terms of their associated benefits and risks to humans, other organisms and the environment	
	b) Discuss the risk associated with an activity in terms of the actual level of the risk and its potential consequences, associated uncertainties, and the factors affecting people's perception of the risk	
10) Consider ethical issues in the treatment of humans, other organisms and the environment	 a) Identify ethical issues arising from the application of science as it impacts on humans, other organisms and the environment b) Discuss scientific solutions 	
11) Appreciate the role of the scientific community in validating new knowledge and ensuring integrity	from a range of ethical viewpoints a) Discuss the importance of critical evaluation of new data or new interpretations of data which challenge established scientific theories or propose new theories	
	b) Describe how the process of communication through journals and conferences and peer reviews contributes to the validation of new scientific theories by the scientific community	
12) Appreciate the ways in which society uses science to inform decision-making	Discuss how science influences decisions on an individual, local, national or international level	

Temperature affects the rate of enzyme controlled reactions. Remind yourself how by looking this work up (your AS notes will help) and then filling in the spaces below:

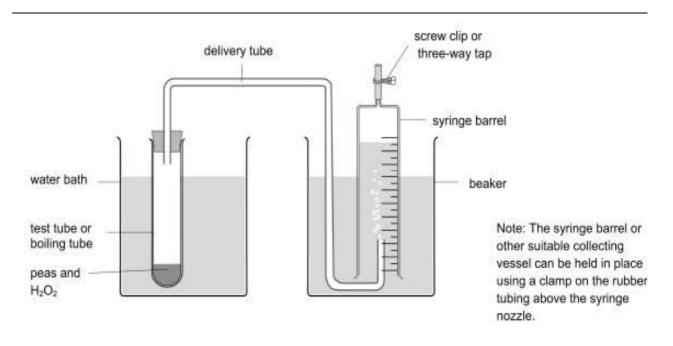


In this experiment we will investigate the effect of temperature on the activity of enzymes in potatoes. The idea is that in order for the potato to develop properly, the enzymes inside it need to be at the right temperature. We will examinine how the activity of the enzyme **catalase** (inside potato cells) is effected by temperature.

$$\begin{array}{rcl} & \text{Catalase} \\ 2H_2O_2 & \longrightarrow & O_2 & + & 2H_2O \end{array}$$

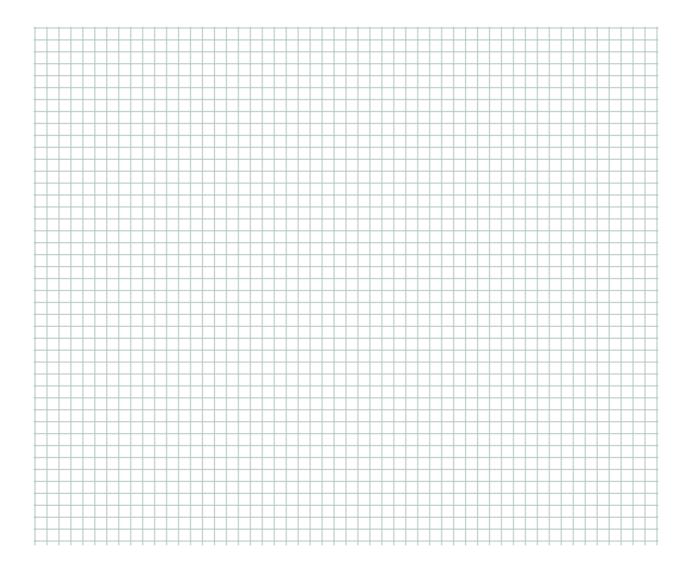
<u>Method:</u>

- 1. Cut a cylinder of potato using a cork borer
- 2. Cut the cylinder to a standard length (e.g. 2cm)
- 3. Set up a boiling tube, delivery tube and gas syringe as shown in the diagram below



- 4. Place the boiling tube in a waterbath at 20°C to acclimatise
- 5. Add the potato cylinder to the boiling tube
- 6. Add 5cm3 of H₂O₂ solution (20vol conc)
- 7. Record the cumulative volume of gas emitted every 15s for 3min
- 8. Repeat the process for the same temperature and calculate the mean gas emitted for each interval
- 9. Repeat the experiment for a range of different temperatures
- 10. Use your data to fill in the table and plot a suitable graph

		Cumulative volume of gas emitted per time / cm ³											
Temperature	0s	15s	30s	45s	60s	75s	90s	105s	120s	135s	150s	165s	180s
1													
2													
Av													
1													
2													
Av													
1													
2													
Av													
1													
2													
Av													
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<u>Trends:</u>

Explanation of the trends:

Questions

1. What risks were present in this experiment?

2. What implications does this have for germination of seeds?

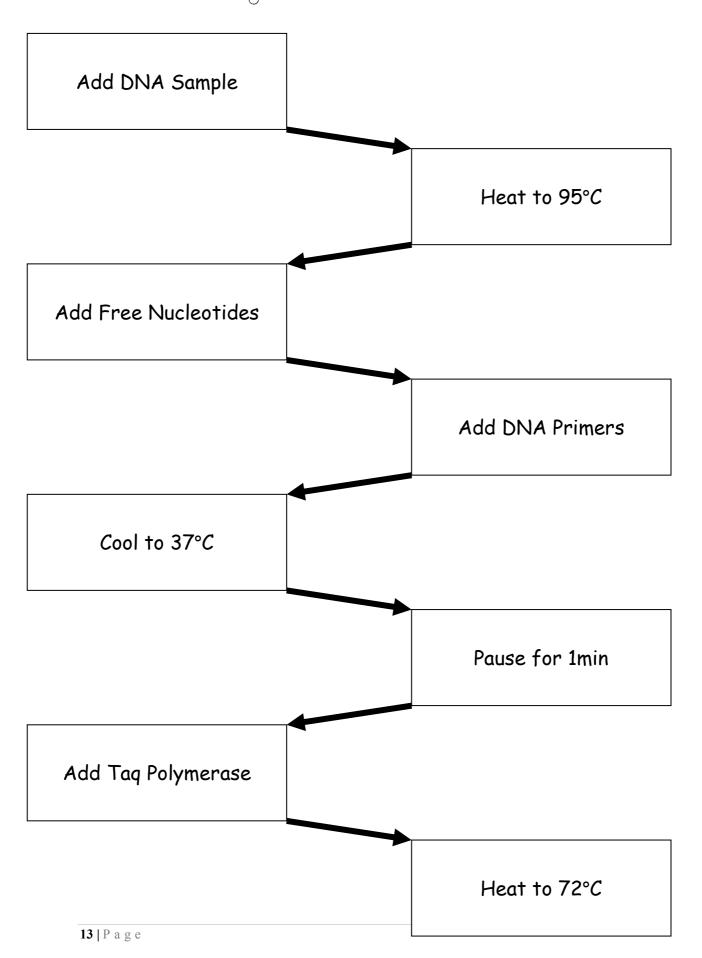
3. What impact might global warming have on this?

4. What other organisms' development is affected by temperature?

5. What second organism are you specifically asked about in your syllabus?

Extension 1: Are all organisms adapted to have an optimum temp at $\sim 40^{\circ}C$?

6.6 Polymerase Chain Reaction (PCR)



Questions

1. What is the purpose of PCR?

2. When is PCR commonly used?

3. Why heat to 95°C?

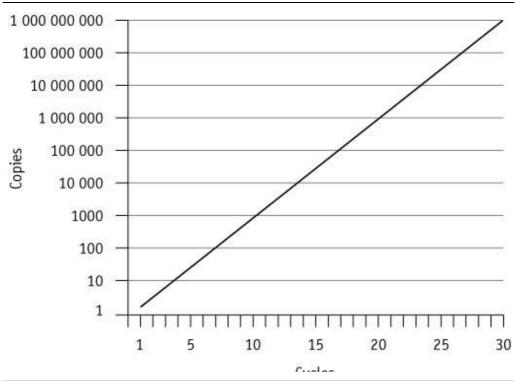
4. What are the primers for (and what's a primer)?

5. Why cool to 37°C?

6. Why heat to 72°C?

The Forensic Science Service use PCR to produce millions of copies of the STR fragments used in producing a DNA profile. Work through the interactive tutorial on the polymerase chain reaction (PCR) which accompanies this activity and use the A2 textbook to help you complete the following exercise.

- 1. The enzyme *Taq Polymerase* is only added during the first PCR cycle, but continues to catalyse DNA replication through many cycles. Considering the treatment of the DNA during PCR, what property does polymerase show that is unusual for an enzyme?
- 2. Where does Tag Polymerase come from?
- 3. What feature of a DNA molecule ensures accurate replication of the strands during each PCR cycle?
- 4. Explain how PCR has revolutionised criminal investigations.
- 5. The graph in Figure 1 shows the potential number of copies of DNA produced during PCR. Use your understanding of the process to explain how so many copies can be produced in relatively few cycles.



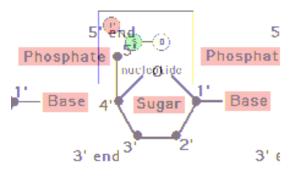
Use this page for your answers:

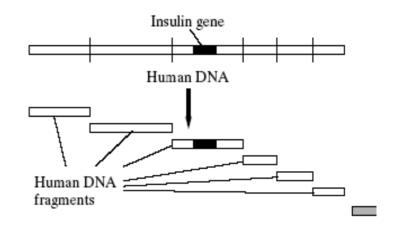
6.7 Electrophoresis

Electrophoresis is the technique used to separate lots of pieces of DNA of different sizes.

For example, if we wish to cut out a gene from a length of DNA we use a restriction enzyme. However, the restriction enzyme may cut the length of DNA in lots of places, producing lots of pieces of DNA. How can we select the piece with the gene in?

Answer: Use electrophoresis



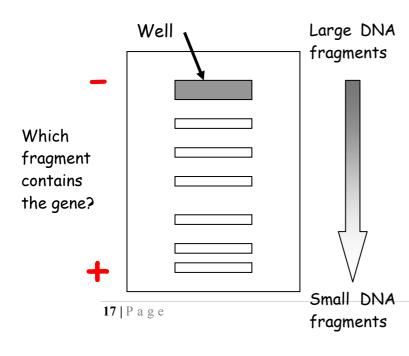


DNA is made from nucleotides joined together by the sugar- phosphate backbone.

Each nucleotide contains;

leotide

The phosphate group (PO_4^{2-}) is strongly negatively charged. This means DNA will be repulsed from negatively charged things. This is the central idea between electrophoresis.



Pieces of DNA are placed in a well (hole) at one end of a sheet of agar jelly. A strong current is passed through the agar (the well is next to the negative electrode)

The DNA is repulsed from the negative electrode and begins to move away through the agar fibres. Small pieces of DNA can move quickly through the fibres and move further. Large pieces get stuck easily & don't get far.

Questions

1. What is a restriction enzyme?

2. What is the purpose of electrophoresis?

3. Where is electrophoresis used commonly?

4. When are gene probes used in electrophoresis?

5. What is Southern Blotting?

6. What techniques are available to make DNA visible?

Electrophoresis Kit:

You may well use an electrophoresis kit to demonstrate the theory on the previous pages. If you do, use these pages to take notes and answer any questions set by your teacher.

6.18 Which antibiotic is the most effective?

When a bacterial infection is diagnosed antibiotics may be prescribed. Different antibiotics are not equally effective against all bacteria, so the correct antibiotic must be selected for a particular bacterial infection. In some cases the most effective antibiotic is known, but in other cases tests need to be carried out by a pathology department. In this activity you will be testing the effectiveness of several types of antibiotics on bacteria.

The standard method of doing this is to put discs of blotting paper soaked in the various antibiotics onto an agar plate that has been inoculated with the bacteria. Alternatively a mast ring (a ring of paper with several 'arms', each treated with a different antibiotic) can be used.

You need

- Agar plate seeded with known bacteria
- Sterile Pasteur pipette
- Bunsen burner
- Beaker of disinfectant, 1% Virkon or equivalent
- Bench spray of disinfectant, 1% Virkon or equivalent
- Bactericidal soap
- Paper towels
- Marker pen
- Forceps
- Mast ring or antibioticimpregnated paper discs
- Adhesive tape
- Incubator set at 30 °C

Safety

Wear eye protection.

The microorganisms are a potential biological hazard. Use aseptic techniques when transferring the bacteria to the Petri dishes. Clean the bench with antibacterial disinfectant. Do NOT open the Petri dishes once they have been incubated.



Procedure

- 1 Wash your hands with the bactericidal soap. Spray the working area thoroughly with the disinfectant spray and wipe with a paper towel after waiting for the disinfectant to act (10 minutes with Virkon, longer with other disinfectants).
- 2 Prepare an agar plate seeded with bacteria. This may have already been done for you. Label the Petri dish on the base at the edge with your name, the date, and the type of bacterium it is inoculated with.

- 3 Sterilise the forceps by flaming them and allow to cool. Use them to pick up an antibiotic disc or mast ring. Raise the lid of the Petri dish and place the mast ring firmly in the centre of the agar; if individual discs are used they will need to be spaced evenly around the dish.
- 4 Tape the dish securely with two pieces of adhesive tape (but do not seal it completely), then incubate it upside down for 48 hours at 30 °C.
- 5 Wash your hands with bactericidal soap and clean the bench again using the Virkon spray.
- 6 After incubation, look carefully at the plate but do not open it. Where bacteria have grown, the plate will look opaque, but where the antibiotics have inhibited growth, clear areas called inhibition zones will be seen. Measure the diameter of the inhibition zones in millimetres and use this information to decide which antibiotic is most effective at inhibiting the growth of the bacterium.
- 7 Collect data from other members of the class who used the other bacterial cultures.
- 8 Complete the table overleaf by collating the class data

Table:

	Diameter of Ring / mm						
Antibiotic	1	2	3	4	5	6	Average

Questions

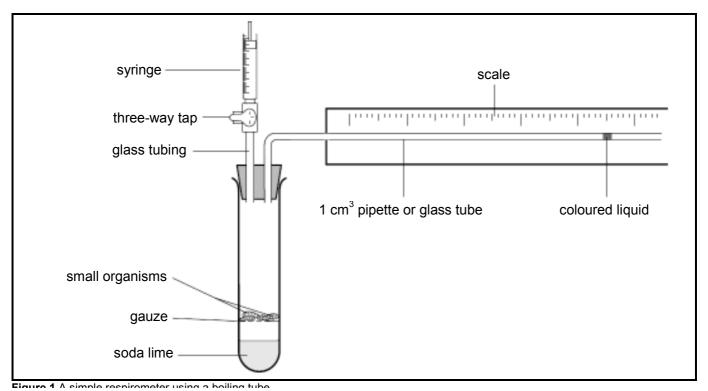
1 What factors determine the diameter of the inhibition zones?

2 Why were you told to incubate the plates at 30°C when human body temperature is 37°C?

3 If you were working in a hospital laboratory, and you had just carried out this test on bacteria isolated from sick patients, would you always choose the antibiotic that gave the biggest inhibition zone? Are there any other factors you would need to consider?

7.6 Investigating the rate of respiration

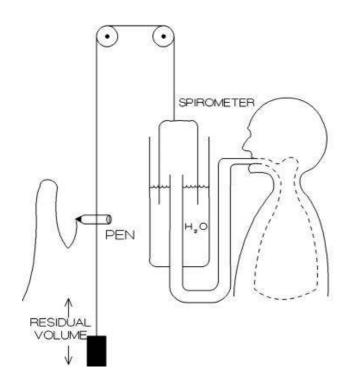
The rate of respiration is proportional to the volume of O_2 used per unit time. This can be measured using a simple **respirometer** (see below)



However, this kind of respirometer leaks frequently and is notoriously difficult to get useful data from. Therefore, we are going to do a similar experiment using a **spirometer**.

The spirometer is essentially a fixed volume chamber floating in a bath of water. Every time we breathe in the total volume of the chamber decreases and every time we breathe out the volume increases. By attaching a pen to the chamber, we can get a qualitative recording of the volume of air in our lungs at any time.

For safety reasons the chamber is filled with pure O_2 . Every time we inhale we breathe in 100% O_2 and then breathe out ~95% O_2 and ~5% CO_2 . CO_2 is poisonous in high concentration, so the CO2 is removed from the exhaled air by a soda lime scrubber. This means that the total volume of the spirometer falls slowly in proportion to the volume of O_2 used per unit time.



A simple diagram of a spirometer

Some Definitions:

Total Lung Capacity = Vital Capacity + Residual Volume

Total Lung Capacity:

Vital Capacity:

Residual Volume:

Before we can record the volume of O2 used per minute we need to **calibrate** the spirometer. What does this means and why do we do it?

Stick a copy of the spirometer chart here and use it to answer the questions on the next page

Questions

1. How many small vertical squares represent one litre (use the initial calibrations)?

2. How many small horizontal squares represent one minute (use the initial calibrations)?

3. By calculating the gradient of the spirometer chart, give the metabolic consumption of oxygen per minute?

4. Calculate the minute volume for the first minute of the recording

5. How might the spirometer chart be different for a long-term smoker?

The following spirometer trace was produced for a student at rest (bottom line) and during exercise (top line)

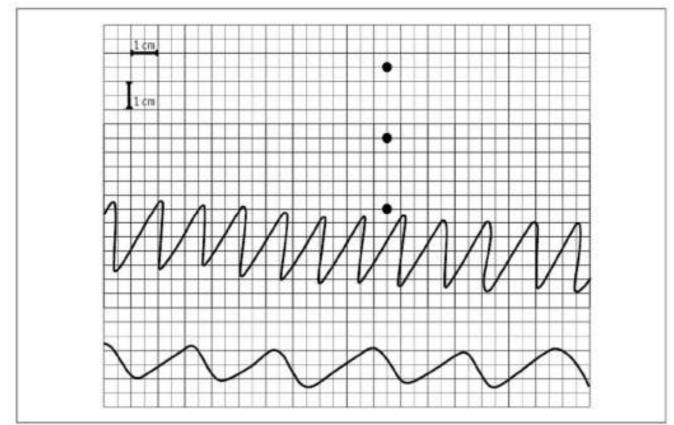


Figure 2 Spirometer chart: top trace = after exercise; bottom trace = before exercise.

The three dots above the upper trace are the calibration dots: the first dot, the lowest of the three, is the baseline level recorded before any oxygen is added, the second dot is after adding 1 dm³ oxygen to the chamber within the spirometer, and the third dot is after adding another 1 dm³ oxygen. The chart recorder was set at 0.5 cm s^{-1} . (NB 1 dm³ is the same as 1 litre; 1 dm³ = 1000 cm³.)

Questions

- 1. Use the trace in the figure to find the effect that exercise had on breathing volume and rate. Suggest and explanation for your findings
- 2. If you were asked to investigate other changes to the body during exercise, what other factors could you measure easily?
- 3. Spirometers are used to calculate a subject's BMR from the amount of oxygen consumed in a given time. Explain why it would be difficult to measure your own BMR in this way during a biology lesson

Extension:

What is the Respiratory Quotient? How is this measure useful to scientists?

8.15 Habituation to a stimulus

Many people will have touched a snail in the garden and noticed that it immediately withdraws its eye stalks. For such a slow-moving animal this seems a very quick response. This suggests that it is important for protection and survival. A snail only withdraws when it is either inactive or threatened. When touched, it withdraws to avoid danger. Do snails become habituated to the stimulus, ceasing to withdraw with repeated stimulation? In this investigation you will collect data to find out if habituation to a touch stimulus does occur in these organisms.

Safety

Wash your hands thoroughly after touching the snails, once all the equipment has been put ready for disinfection. Take care that the stimulus causes no harm to the snails.

Procedure





You need

- One giant African land snail (or a garden snail if not available)
- One dampened cotton wool bud
- A suitable clean, firm surface for the snails (e.g. a plastic chopping board)
- A stopwatch

A

Figure 1 A giant African land snail with eye stalks A extended and B retracted.

R

Method:

- 1. Collect one giant African land snail and place it on a clean, firm surface. Wait for a few minutes until the snail has fully emerged from its shell (Figure 1A) and is used to its new surroundings.
- 2. Dampen a cotton wool bud with water.

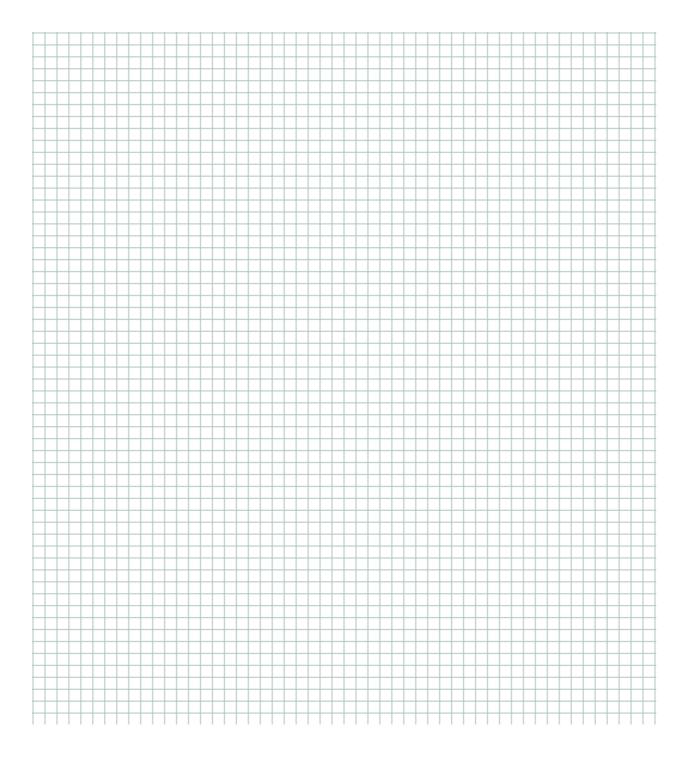


- 3. Firmly touch the snail between the eye stalks with the dampened cotton wool bud and immediately start the stopwatch. Measure the length of time between the touch and the snail being fully emerged from its shell once again, with its eye stalks fully extended.
- 4. Repeat the procedure in step 3 for a total of ten touches, timing how long the snail takes to re-emerge each time.
- 5. Record your results in the table below and plot a suitable graph

<u>Table:</u>

Touch number	Time between touch and the snail being fully emerged / sec
1	
2	
3	
4	
5	
6	
7	
8	
9	
10	

<u>Graph:</u>



Questions

1. Write a hypothesis which this experiment will test

- 2. Using your graph, state whether you think there is a positive, negative or no correlation between the number of stimulations and the time for eye stalk withdrawal.
- 3. Explain any patterns or trends in your data, supporting your ideas with evidence from the data and your biological knowledge of habituation. Relate your findings to your hypothesis.
- 4. Suggest a reason why snails may become habituated to a prodding stimulus in the wild.
- 5. This experiment has been shown to be less successful if the snails are handled regularly prior to the experiment. Suggest why handling prior to the experiment could affect the results of the experiment.

Core Practical Revision

There will <u>definitely</u> be at least one question on each A2 paper about a core practical. Questions tend to fall into two categories;

- 1. Outline a method you could use to test... (i.e. write out the method for a core practical).
- 2. Analyze data from a practical...

The second type of question relies both on your ability to think like a scientist on the spot and also on your working knowledge of the theory behind each practical, so make sure you know exactly why the IV causes the DV to change as it does.

The first type of question is more difficult. However, there are always marks available for the following;

- 1. Specific Procedure or <u>M</u>ethod
- 2. Specific Equipment
- 3. Safety & <u>R</u>isk
- 4. **C**ontrolled Factors you kept constant
- 5. A <u>C</u>ontrol for comparison
- 6. Repeats & taking an <u>A</u>verage
- 7. Stated <u>R</u>ange

One way of remembering this is the mnemonic MERC CAR, like this one \fi



As you progress through the year try why not fill out the blank revision cards below? That way you have a complete record of what you need to learn in the summer *before* the summer!

One final comment for you to think about:

If you miss any of the Core Practicals this year it is <u>imperative</u> that you catch up the work you have missed. Arguably, they are more important than the theory you complete in class because you know for certain that <u>at</u> <u>least one practical will appear on each paper</u>, so each practical has (in theory) ~ 1 in 4 chance of coming up.

See... important!

5.11 - How to study the ecology of an area (see coursework)
Method:
Equipment:
Risks:
Controlled factors:
Control for comparison:
Average:
Range:
Additional Notes:

5.17 - How temperature affects the development of organisms
Method:
Equipment:
Risks:
Controlled factors:
Control for comparison:
Average:
Range:
Additional Notes:

6.6 - Polymerase Chain Reaction (PCR)

This practical is about procedure, so make notes here about what you did and why you did it...

Notes:

6.7 - Electrophoresis

This practical is about procedure, so make notes here about what you did and why you did it...

Notes:

6.18 -	Which antibiotic is the most effective?
Method:	
Equipment:	
Risks:	
Controlled factors:	none
Control for comparison:	none
Average:	
Range:	none
Additional Notes:	

	7.6 - Investigating the rate of respiration
Method:	
Equipment:	
Risks:	
Controlled factors:	
Control for comparison:	
Average:	
Range:	
Additional Notes:	

7.14 - Spirometer and exercise

This practical is about procedure, so make notes here about what you did and why you did it...

Notes:

	8.15 - Habituation to a stimulus
Method:	
Equipment:	
Risks:	
Controlled factors:	
Control for comparison:	
Average:	
Range:	
Additional Notes:	