

AQA A2 Biology Unit 4

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I would be interested to hear of any comments and corrections.

Neil C Millar (nmillar@ntlworld.co.uk)
Head of Biology, Heckmondwike Grammar School
High Street, Heckmondwike, WF16 0AH
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Biology Unit 4 Specification

Ecology

Fieldwork

A critical appreciation of some of the ways in which the numbers and distribution of organisms may be investigated.

- Random sampling with quadrats and counting along transects to obtain quantitative data.
- The use of percentage cover and frequency as measures of abundance.
- The use of mark-release-recapture for more mobile species.

Carry out fieldwork involving the use of frame quadrats and line transects, and the measurement of a specific abiotic factor. Collect quantitative data investigating populations from at least one habitat, including appropriate risk management. Consider ethical issues arising when carrying out field work, particularly those relating to the organisms involved and their environment.

Statistics

Analyse and interpret data relating to the distribution of organisms, recognising correlations and causal relationships. Apply elementary statistical analysis to the results. Appreciate the tentative nature of conclusions that may be drawn from such data.

Populations and the Niche

A population is all the organisms of one species in a habitat. Populations of different species form a community. Population size may vary as a result of the effect of abiotic factors and interactions between organisms: interspecific and intraspecific competition and predation. Within a habitat a species occupies a niche governed by adaptation to both biotic and abiotic conditions.

Succession

Succession from pioneer species to climax community. At each stage in succession certain species may be recognised that change the environment so that it becomes more suitable for other species. The changes in the abiotic environment result in a less hostile environment and changing diversity.

Conservation

Conservation of habitats frequently involves management of succession. Present scientific arguments and ideas relating to the conservation of species and habitats. Evaluate evidence and data concerning issues relating to the conservation of species and habitats and consider conflicting evidence. Explain how conservation relies on science to inform decision making.

Nutrient Cycles

The role of microorganisms in the carbon and nitrogen cycles in sufficient detail to illustrate the

processes of saprobial nutrition, ammonification, nitrification, nitrogen fixation and denitrification. (The names of individual species are not required.)

Energy Flow

Photosynthesis is the main route by which energy enters an ecosystem. Energy is transferred through the trophic levels in food chains and food webs and is dissipated. Quantitative consideration of the efficiency of energy transfer between trophic levels. Pyramids of numbers, biomass and energy and their relationship to their corresponding food chains and webs. Net productivity as defined by the expression

$$\text{Net productivity} = \text{Gross productivity} - \text{Respiratory loss}$$

Intensive Farming

Comparison of natural ecosystems and those based on modern intensive farming in terms of energy input and productivity. The ways in which productivity is affected by farming practices that increase the efficiency of energy conversion. These include

- the use of natural and artificial fertilisers
- The environmental issues arising from the use of fertilisers. Leaching and eutrophication. Analyse, interpret and evaluate data relating to eutrophication.
- the use of chemical pesticides, biological agents and integrated systems in controlling pests on agricultural crops
- intensive rearing of domestic livestock.

Apply understanding of biological principles to present scientific arguments that explain how these and other farming practices affect productivity. Evaluate economic and environmental issues involved with farming practices that increase productivity. Consider ethical issues arising from enhancement of productivity.

Greenhouse Effect

The importance of respiration, photosynthesis and human activity in giving rise to short-term fluctuation and long-term change in global carbon dioxide concentration. The roles of carbon dioxide and methane in enhancing the greenhouse effect and bringing about global warming. Analyse, interpret and evaluate data relating to evidence of global warming and its effects on the yield of crop plants; the life-cycles and numbers of insect pests; and the distribution and numbers of wild animals and plants.

Human populations

Population size and structure, population growth rate, age population pyramids, survival rates and life expectancy. Interpret growth curves, survival curves and age pyramids. Calculate population growth rates from data on birth rate and death rate. Relate changes in the size and structure of human populations to different stages in demographic transition.

Metabolism

The synthesis of ATP from ADP and phosphate and its role as the immediate source of energy for biological processes.

Aerobic respiration

Aerobic respiration in such detail as to show that

- Glycolysis takes place in the cytoplasm and involves the oxidation of glucose to pyruvate with a net gain of ATP and reduced NAD
- Pyruvate combines with coenzyme A in the link reaction to produce acetylcoenzyme A
- Acetylcoenzyme A is effectively a two carbon molecule that combines with a four carbon molecule to produce a six carbon molecule which enters the Krebs cycle. In a series of oxidation-reduction reactions the Krebs cycle generates reduced coenzymes and ATP by substrate-level phosphorylation, and carbon dioxide is lost.
- Synthesis of ATP is associated with the transfer of electrons down the electron transport chain and passage of protons across mitochondrial membranes.

Investigate the effect of a specific variable such as substrate or temperature on the rate of respiration of a suitable organism.

Anaerobic respiration

Glycolysis followed by the production of ethanol or lactate and the regeneration of NAD in anaerobic respiration.

Photosynthesis

The light-independent and light-dependent reactions in a typical C₃ plant.

- The light-dependent reaction in such detail as to show that: light energy excites electrons in chlorophyll; energy from these excited electrons generates ATP and reduced NADP; the production of ATP involves electron transfer associated with the electron transfer chain in chloroplast membranes; photolysis of water produces protons, electrons and oxygen.
- The light-independent reaction in such detail as to show that: carbon dioxide is accepted by ribulose biphosphate (RuBP) to form two molecules of glycerate 3-phosphate (GP); ATP and reduced NADP are required for the reduction of GP to triose phosphate; RuBP is regenerated in the Calvin cycle; Triose phosphate is converted to useful organic substances.

Limiting Factors

The principle of limiting factors as applied to the effects of temperature, carbon dioxide concentration

and light intensity on the rate of photosynthesis. Investigate the effect of a specific limiting factor such as light intensity, carbon dioxide concentration or temperature on the rate of photosynthesis. Candidates should be able to explain how growers apply a knowledge of limiting factors in enhancing temperature, carbon dioxide concentration and light intensity in commercial glasshouses. They should also be able to evaluate such applications using appropriate data.

Genetics

Genetic Crosses

The genotype is the genetic constitution of an organism. The phenotype is the expression of this genetic constitution and its interaction with the environment. The alleles at a specific locus may be either homozygous or heterozygous. Alleles may be dominant, recessive or codominant. There may be multiple alleles of a single gene. Use fully labelled genetic diagrams to predict the results of

- monohybrid crosses involving dominant, recessive and codominant alleles
- crosses involving multiple alleles and sex-linked characteristics.

The Hardy-Weinberg Principle

Species exist as one or more populations. The concepts of gene pool and allele frequency.

- Calculate allele, genotype and phenotype frequencies from appropriate data and from the Hardy-Weinberg equation, $p^2 + 2pq + q^2 = 1$ where p is the frequency of the dominant allele and q is the frequency of the recessive allele.
- The Hardy-Weinberg principle. The conditions under which the principle applies. Understand that the Hardy-Weinberg principle provides a mathematical model that predicts that allele frequencies will not change from generation to generation.

Natural Selection

Differential reproductive success and its effect on the allele frequency within a gene pool. Directional and stabilising selection. Use both specific examples and unfamiliar information to explain how selection produces changes within a species. Interpret data relating to the effect of selection in producing change within populations.

Speciation

Geographic separation of populations of a species can result in the accumulation of difference in the gene pools. The importance of geographic isolation in the formation of new species.

Ecology

Ecology (or environmental biology) is the study of living organisms and their environment. Its aim is to explain why organisms live where they do. To do this ecologists study ecosystems: areas that can vary in size from a pond to the whole planet.

Biosphere	The part of the planet Earth where life occurs, including land, sea and air.
Ecosystem	A reasonably self-contained area together with all its living organisms, e.g. oak forest, deep sea, sand dune, rocky shore, moorland, hedgerow, garden pond, etc.
Habitat	The physical or <u>abiotic</u> part of an ecosystem, i.e. a defined area with specific characteristics where the organisms live. Most ecosystems have several habitats.
Microhabitat	A localised specific habitat within a larger habitat e.g. under a rotting log, in a rock pool, etc.
Terrestrial	An ecosystem on dry land
Aquatic	An ecosystem in water
Marine	An ecosystem in the sea
Community	The living or <u>biotic</u> part of an ecosystem, i.e. <u>all</u> the organisms of <u>all</u> the different species living in one habitat.
Biotic	Any living or biological factor.
Abiotic	Any non-living or physical factor.
Population	The members of the <u>same species</u> living in one habitat.
Species	A group of organisms that can successfully interbreed

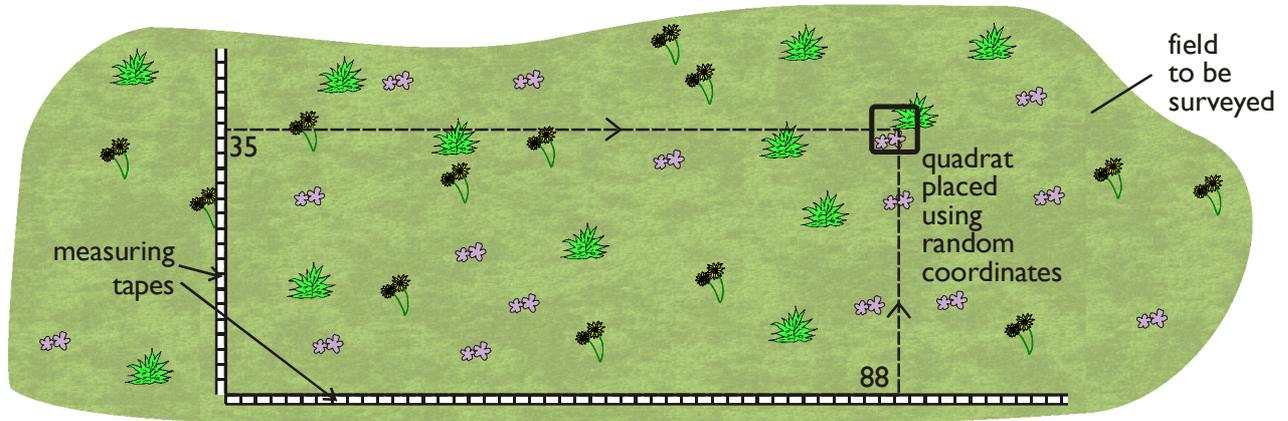
Fieldwork

Ecology is best studied in the organisms' natural habitat, but working in "the field" presents particular practical difficulties: the habitats can be very large; there can be a very large number of different organisms present; many of the organisms move about or are difficult to find; the organisms can be difficult to identify; some organisms may eat other organisms; and confounding variables, like the weather, can be impossible to control. To deal with these problems there are a number of specific fieldwork techniques.

Sampling

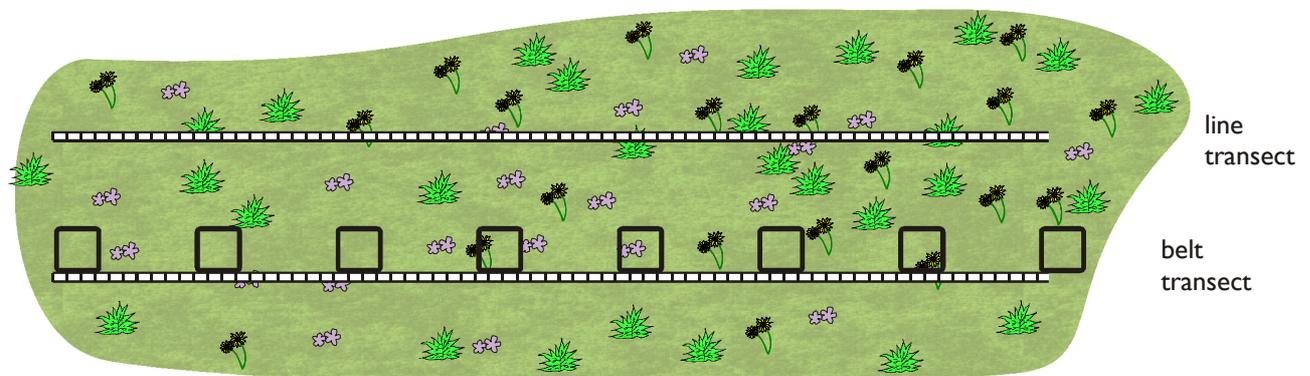
In unit 2 we came across the idea of sampling a population, in other words looking at a small sample of the biota in an ecosystem, rather than studying every living thing, which would be impossible. There are two strategies for sampling an ecosystem, depending on your objective.

- **Random sampling** is used when you want a representative sample of the whole area under study. Measuring tapes are placed along two sides of the area, like axes of a graph, and random numbers (from tables or a computer) are used as coordinates to choose sampling points in the area. Alternatively, random numbers can be used as polar coordinates (angle and distance) starting from a central point. Walking aimlessly, throwing things and choosing sites are not random methods!

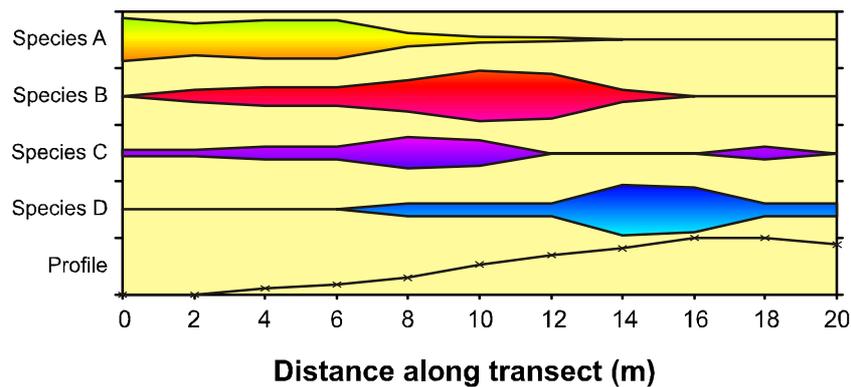


There should always be a large number of samples (at least 10, and preferably 100) to minimise the chance of picking a skewed sample and to allow for bad measurements or anomalies. One should aim to sample at least 2% of the total area, so if the field area was 500m², you would need to sample 10 m² of the area altogether.

- **Systematic sampling** is used when you choose where to take your samples, because you are investigating a specific pattern in the ecosystem. The most common kind of systematic sample is a transect, where samples are taken along a straight line to see what changes there are along the line. The transect usually follows an environmental gradient, such as down a rocky shore, into a forest or down a mountain side. The transect could be a few metres long or a few 100 km long. In a line transect the organisms touching a piece of string stretched along the transect are recorded. In a belt transect quadrats are placed at intervals along the transect and organisms in each quadrat are counted. The line transect is quick but can be unrepresentative while the belt transect involves more work, but can generate more complete data.



The data from a transect can be presented as a kite graph, which shows biotic data as “kites” and abiotic data as lines:



However the sampling sites are chosen, both biotic and abiotic factors should be measured at each sampling site. The combination of the two measurements gives a more detailed understanding of the ecosystem. We'll now look at specific techniques for measuring abiotic and biotic factors.

Measuring Abiotic Factors

Abiotic factors in ecology are usually measured with special digital electronic equipment. The electronic devices usually consist of a sensor or probe (such as a temperature probe, pH probe or light sensor) connected to an amplifier and digital display. These devices have many advantages: the measurements are quick, quantitative, accurate, calibrated, and can be automatically recorded at regular time intervals over an extended period of time. The data can also be transferred to computers (wirelessly if necessary) for storage and analysis. Some measurements (like soil depth) are still best done with conventional equipment (like a ruler). Almost any abiotic factor can be measured:

- In an aquatic habitat you might measure the water temperature; the oxygen concentration (usually as percent saturation to allow for changes due to temperature); water pH, turbidity (which measures suspended solids); conductivity (which measures total dissolved ions); specific mineral concentrations (using chemical tests); flow rate; etc.
- In a terrestrial habitat you might measure soil (edaphic) properties, such as soil temperature; soil pH; soil moisture; soil depth; soil texture; soil composition; etc.
- In a terrestrial habitat you might measure air temperature; light intensity; wind speed and direction; air humidity; etc.
- On a slope you might measure altitude; slope gradient; slope aspect (direction); profile; etc.

As with any measurement, each abiotic measurement should be repeated several times at each sampling site and averaged. The measurements might also be repeated over the course of a day or a year, to account for daily and seasonal variations. Generally the aim will be to correlate the abiotic measurements with biotic measurements taken at the same points, to see if there might be a causal relationship, i.e. the abiotic factors could explain the distributions of the living organisms.

Measuring Biotic Factors

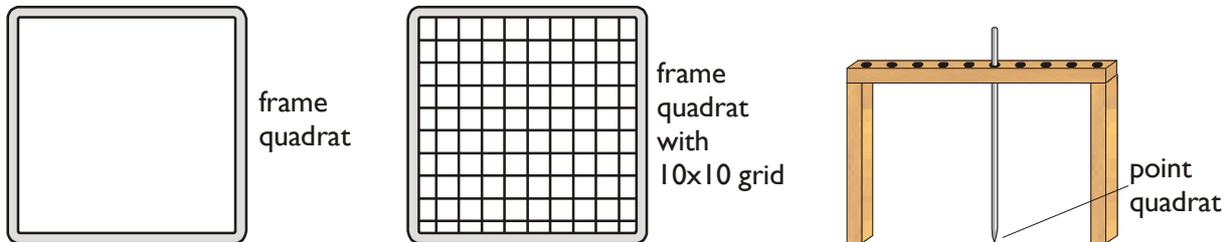
How can we measure the living organisms in our sample sites? The first step is to find them and then to identify them. Techniques for finding plants and animals are listed below, and identification is carried out using identification keys, which allow organisms to be identified using simple questions about their appearance. Sometimes we want to include all the living organisms in our samples, but often we are only interested in certain groups (like plants or invertebrates) or even just one species. There are many different quantitative measurements one can make of living organisms, depending on the purpose of the investigation:

- **Abundance.** The most obvious measurement is simply to count the number of organisms in the sample. This measurement is called the abundance. Usually we identify each organism found and so record the abundance of each species, but sometimes we simply count total abundance of all species. For animals we need to use the capture-mark-recapture method for counting the number of organisms, since they move (see p 13). Often we divide the abundance by the sampling area to calculate the density – the number per square metre. However, it is sometimes impossible to distinguish between closely-spaced individuals, such as with grasses.
- **Richness.** This is the number of different species found in the sample. It is a simple measure of diversity.
- **Diversity.** As we saw in unit 2, a better measure of diversity is the Simpson Diversity Index (D), which takes into account the species richness and their abundance. Its formula was given in unit 2.
- **Growth.** Sometimes we are interested in comparing the growth or size of similar organisms in different habitats. For animals this might be done by measuring their mean length, or wing span, or recording their mass. For plants this might be done by measuring mean plant height, leaf area, number of leaves or plant mass (though this would mean uprooting and killing the plant).
- **Biomass.** For studying productivity and making pyramids of biomass we need to measure dry mass, since most of a living organism's mass is made of water, which doesn't contain energy. To obtain the dry mass a sample of the organisms must be warmed in an oven at about 80°C to evaporate the water, but not burn any organic material. The sample is weighed at intervals until the mass no longer decreases, because all the water has been evaporated. This technique is drying to constant mass.

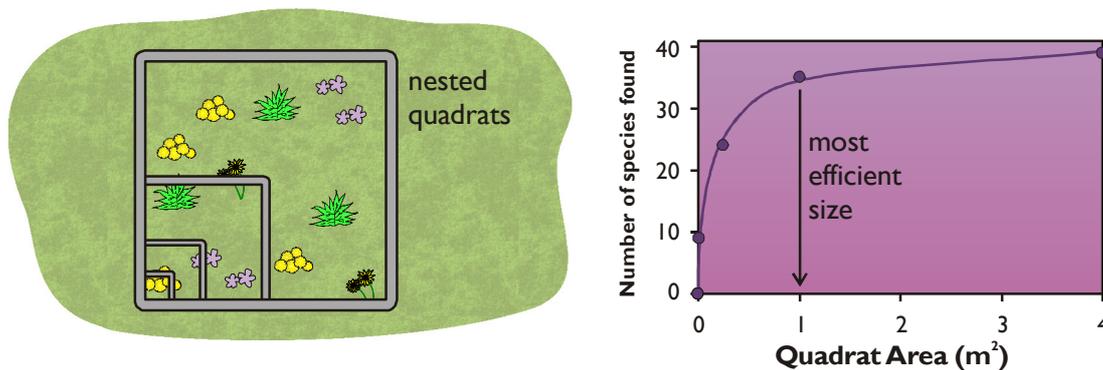
We'll now look at specific techniques for sampling plants and animals.

Sampling Plants

Plants are most easily sampled using a quadrat, since they don't move. Quadrats (or frame quadrats) are square frames that are placed on the ground to provide a small, standard area for investigation. Quadrats come in a variety of sizes, commonly 10cm, 50cm or 100cm a side, and may be subdivided into 25 or 100 smaller squares. The smallest quadrat is the point quadrat (or pin quadrat), which is a needle (like a knitting needle), with the point of the needle being the actual tiny quadrat.



A 50cm square is suitable for small plants, grassland and general school work, while a smaller 10cm square would be better for examining lichens on a tree trunk, and a large 1m quadrat would be better in a wood. To find the best size of frame quadrat for a particular habitat, one needs to do a preliminary experiment “nesting” different-sized quadrats in the area to be studied and counting the number of species found. From the species-area graph we can choose a quadrat size that is likely to catch all the species, but without wasting unnecessary effort.



Quadrats allow us to make quantitative measurements of the abundance of plants. There are different ways to do this.

- **Density.** Count the number of individuals of each species in a quadrat, then divide by the area of the quadrat. For example if there is an average of 12 limpets per 0.25m² quadrat, the density is $12/0.25 = 48$ limpets m⁻². This measure isn't appropriate when individual plants are difficult to identify.
- **Species Frequency.** Record the number of quadrats in which a species was found (its frequency). For example if a species was found in 12 quadrats out of a total of 40, then the frequency is $12/40 = 30\%$. Alternatively a quadrat divided into a grid of 25 or 100 smaller squares can be used for plants that are densely-packed. The number of small squares in which the species is found is recorded.
- **Percent Cover.** This is appropriate when it is difficult to identify individual plants (such as grasses). The percentage area of the quadrat covered by that particular species is estimated (to the nearest 5%). This

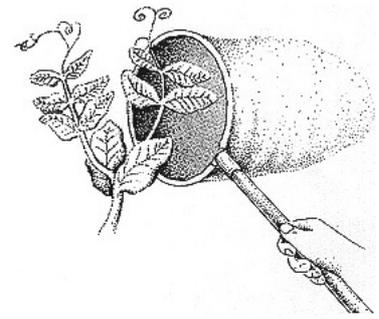
is often easier if the quadrat is subdivided into 25 or 100 smaller squares, but even so it is quite subjective. Since plants can be layered the total percentage cover can be more than 100%.

- Point Quadrats provide an alternative way to measure percent cover. The needle is dropped through a hole in a frame till it touches the ground and whatever species the needle hits are recorded. There are usually 10 holes in one frame to allow for 10 repeats, and then the frame is moved to a large number of other sites to obtain at least 100 repeats. The number of hits divided by the total number of repeats gives the percent cover. For example if a species was hit 66 times out of a total of 200 needles then the percent cover is $66/200 = 33\%$. This is less subjective than using a frame quadrat, and can be very quick with practice.
- Abundance Scale. This is a qualitative way to assess abundance. A common scale is the five-point "ACFOR" scale where A = Abundant; C = Common; F = Frequent; O = Occasional; R = Rare. With practice, this is a very quick way to collect data, but it is not quantitative. The scale can be made semi-quantitative by making the points correspond to ranges of percent cover.

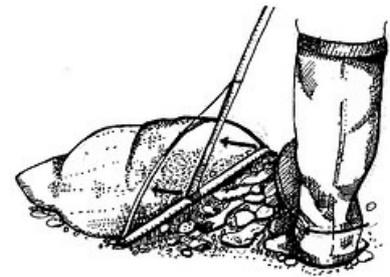
Sampling Animals

Different techniques are needed for sampling animals, since they move. A few animals are sessile and don't move (e.g. limpets and barnacles on a sea shore), while others are sedentary and move only slowly (e.g. snails, woodlice). These animals can be sampled successfully using quadrats. But most animals move too quickly, so need to be caught using nets or traps. There is an almost endless choice of trapping techniques, depending on the particular animals being investigated and their environment, and here is a short selection:

- Sweep nets are large, fine-meshed nets used for insects and other invertebrates living on and around vegetation, especially in grassland and crops. The net is swept back and forth over the vegetation, catching the animals in the process. The animals are then emptied into white trays for identification and counting. The sweeping technique should be standardised (e.g. time, height) to allow fair comparisons between different sites.



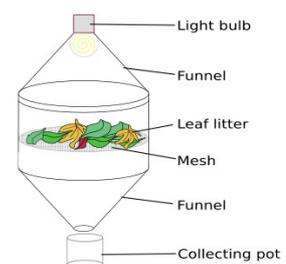
- D-nets are nets with a flat side that can sit on the bed of a stream and are used to catch small aquatic animals such as insect larvae and nymphs. Since these animals are usually well-adapted for burrowing or clinging to rocks they won't be caught unless they are disturbed. So the D-net is held facing upstream and the mud and stones upstream are kicked so that the animals are dislodged and are carried downstream into the net. This technique is called kick sampling. The kick sampling technique should be standardised (e.g. time, area kicked) to allow fair comparisons between different sites.



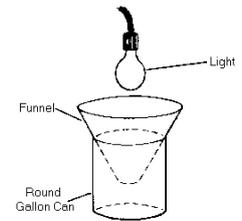
- Beating trays are used for collecting invertebrates from trees and shrubs. The canopy is shaken by hitting with a stick and animals fall into a large white collecting tray or sheet held beneath the branch. The animals can then be captured in a pooter and counted.



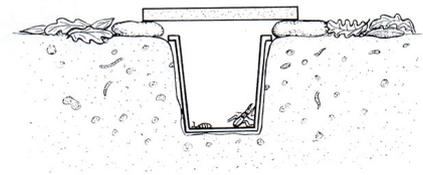
- Tullgren funnels are used to extract invertebrates, particularly arthropods, from samples of leaf litter and soil. The material is placed on a coarse mesh in a large funnel and heated from above with a light bulb. The animals move away from the heat and fall into a collecting vessel, where they can be identified and counted.



- Light Traps are used to catch flying insects, especially at night. Many insects are attracted by the light and fall through the funnel underneath into the trap. Lamps that emit ultraviolet light are good at trapping moths.



- Pitfall traps are used to catch invertebrates that move along the ground, like insect and mites. A smooth-sided cup is buried in the ground with its top level with the surface, and is left overnight. Crawling animals simply fall into the trap and then can't escape up the steep smooth walls. A raised cover keeps out rain and larger animals that might eat the prisoners. The next day the trap is recovered and the animals are recorded and released.



- Longworth traps are used for small mammals like wood mice, shrews and voles. The traps are prepared with dry bedding material and suitable food (such as seeds or fruit), and placed randomly in the area to be surveyed. Small mammals will enter the trap, attracted by the bait, trigger the door and be trapped. They should survive the night in the trap and can be released the next morning. This is particularly important for mammals, as voles are protected by law and can only be trapped with a licence.



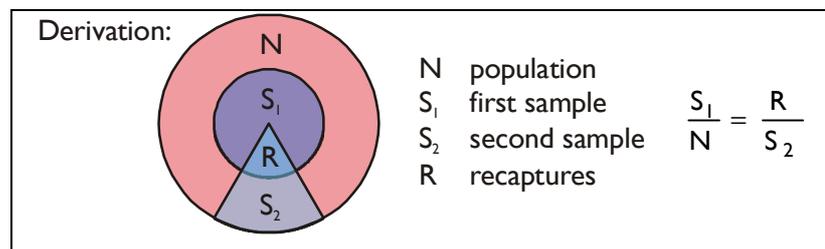
- Sighting methods are used for animals like birds and small mammals. An observer walks (or drives) along a randomly-chosen transect line and counts how many animals, nests, burrows or other evidence he sees. Assuming the animals are distributed randomly in the area one can use the count to estimate the total population.
- Aerial surveys are used for counting large animals over a large area, such as lions in a game reserve. An observer flies over the area either counting all the animals, or taking photographs at random sampling sites, like a huge quadrat.

Capture-Mark-Recapture

Another problem with sampling animals is counting the total number of animals in an area. Unlike plants (or sessile animals), where average density can be measured easily, animals move quickly and often try to remain hidden as much as possible. One solution to these problems is the capture-mark-recapture technique.

- 1 Capture a sample of animals using one of the trapping techniques described above. The larger the sample the better the estimate works.
- 2 Count all the animals in this sample (S_1) and mark them so that they can be recognised later. Typical marks include: a spot of paint for invertebrates, leg-rings for birds, a shaved patch of hair for mammals, small metal disks for fish, etc. Larger animals can also be “marked” by collecting a small blood sample and making a DNA fingerprint (see unit 5).
- 3 Release all the animals where they were caught and give them time to mix with the rest of the population (typically one day).
- 4 Capture a second sample of animals using the same trapping technique.
- 5 Count the animals in the second sample (S_2), and the number of marked (i.e. recaptured) animals in the second sample (R).
- 6 Calculate the population estimate (N , the Lincoln-Petersen Index) using the formula:

$$N = \frac{S_1 S_2}{R}$$



For this formula to be valid three conditions must be met:

- 1 The marking must not affect survival. For example the mark must not make the animal more obvious to predators, or hinder their movement, or harm the animal in any other way. One new solution is to mark with an ultra-violet marking pen (used to check counterfeit notes), which can't be seen under normal sunlight, but can be seen under ultra-violet light.
- 2 The marked animals must have time to mix randomly with the rest of the population before the second sample is taken.
- 3 The population must remain constant between the first and second sampling. In other words there must not be too much time for births, deaths, immigration or emigration to affect the population.

Even when all these criteria are met, the Lincoln-Petersen index is only a very rough estimate of the true population, which is usually in the range $N \pm 50\%$. If the marking is unique for each individual animal (such as numbers on leg-rings) then the marking can also be used to track individual movements, though this is not necessary for calculating the Lincoln-Petersen index.

Analysing Fieldwork using Statistics

Ecological studies often show considerable variation, so it can be difficult to tell whether observed patterns are real or are just due to chance. Suppose we find that mean light intensity is lower in a deciduous wood than in a coniferous wood. Is this difference real, or did we by chance choose darker sites in the deciduous wood? In cases like this an appropriate statistical test can help to clarify the results so that a valid conclusion can be made. The statistical test returns a probability (or P-value), on a scale of 0-1. This P-value is the probability that any observed patterns in the results are just due to chance. So we're hoping for a low P-value: it means that the patterns in the results are probably not due to chance, but instead are significant.

In biology we usually take a probability of 0.05 (5%) as the cut-off. This may seem very low, but it reflects the facts that biology experiments are expected to produce quite varied results. So:

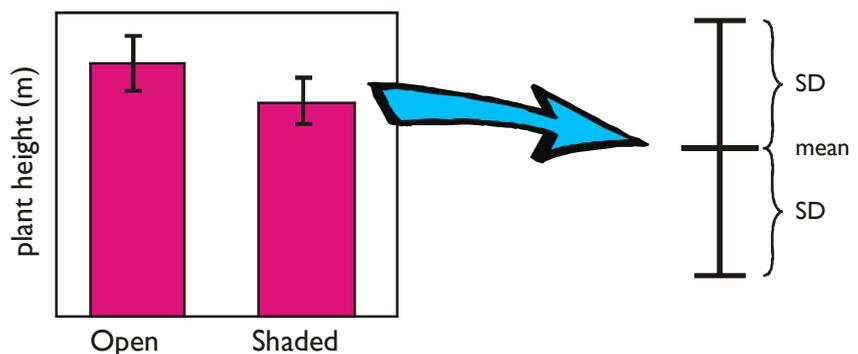
- If $P \geq 0.05$ then we conclude that the observed differences or correlations are just due to chance.
- If $P < 0.05$ then we conclude that there is a significant difference or correlation.

In order to carry out a statistical test we first need make a null hypothesis, so that we are clear about what exactly we're testing for. The null hypothesis always states that there is no difference between groups, or no correlation between variables, and so is fixed for a given investigation. It has nothing to do with (and can be quite different from) any scientific hypothesis you may be making about the result of the experiment. The P-value from the stats test is effectively the probability that the null hypothesis is true. So if $P \geq 0.05$ we accept the null hypothesis, and if $P < 0.05$ then we reject the null hypothesis. Note that the word "significant" is used in the conclusion, but not in the null hypothesis itself.

There are basically three kinds of investigation. We'll look briefly at an example of each in turn.

Looking for Differences (bar chart)

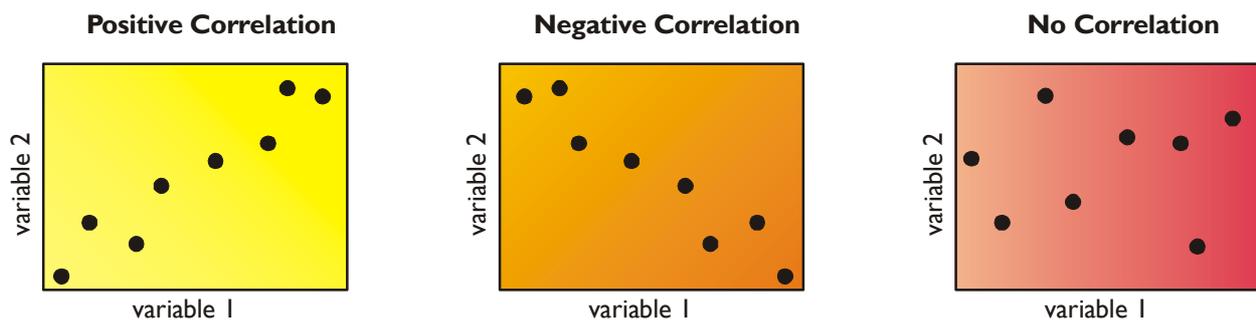
Sometimes we are looking at differences between groups (e.g. are these plants taller than those plants?). We plot a bar chart of the means for each group to see if there is a difference. But how do we know if any differences in mean height are real, or are just due to random chance? We can use the spread of the replicates to find out. In unit 2 we saw that spread can be measured using the standard deviation (SD), but there are other measures of spread, such as the standard error of the mean (SEM) and confidence interval (CI). These spread values



can be added to the bar chart as error bars. If the error bars overlap, then we can say that the observed difference is just due to chance. If they don't overlap, then we can say that there is a significant difference.

Looking for Correlations (scatter graph)

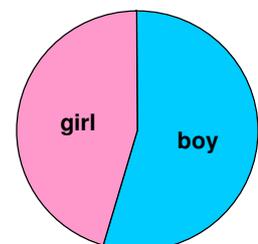
Sometimes we are looking for correlation between two sets of continuous data (e.g. if this goes up does that go up?). We plot a scatter graph of one factor against the other (without a line of best fit) to see if there is a correlation. If both factors increase together then there is a positive correlation; if one factor decreases when the other increases then there is a negative correlation; and if the scatter graph has apparently random points then there is no correlation:



To find the strength of the correlation we calculate a correlation coefficient. It varies from 0 (no correlation) to 1 (perfect correlation). Positive values indicate a positive correlation while negative values indicate a negative correlation. The larger the absolute value (positive or negative), the stronger the correlation (i.e. the closer the data are to a straight line). Remember that a correlation does not necessarily mean that there is a causal relation between the factors (i.e. changes in one factor cause the changes in the other). The changes may both be caused by a third factor, or it could be just coincidence. Further controlled studies would be needed to find out.

Using Qualitative Data (pie chart)

Sometimes we record qualitative (or categoric) data, i.e. observations using words rather than numbers (e.g. colours, shapes, species). It's a little surprising that we can do statistics at all on categoric data, but if a very large number of observations are made then the number of observations of each category can be counted to give frequencies. We plot a pie chart of the observed frequencies and can then compare them with the frequencies expected from a theory. A special statistical test can tell us if the differences between the observed and expected frequencies are significant, or just due to chance. For example the frequencies of boys and girls born in a hospital over a period of time can be compared to an expected 1:1 ratio.



Populations

A population is the number of a particular species living in one habitat. Population Ecology is concerned with the question: why is a population the size it is? This means understanding the various factors that affect the population. Many different factors interact to determine population size, and it can be very difficult to determine which factors are the most important. Factors can be split into two broad groups:

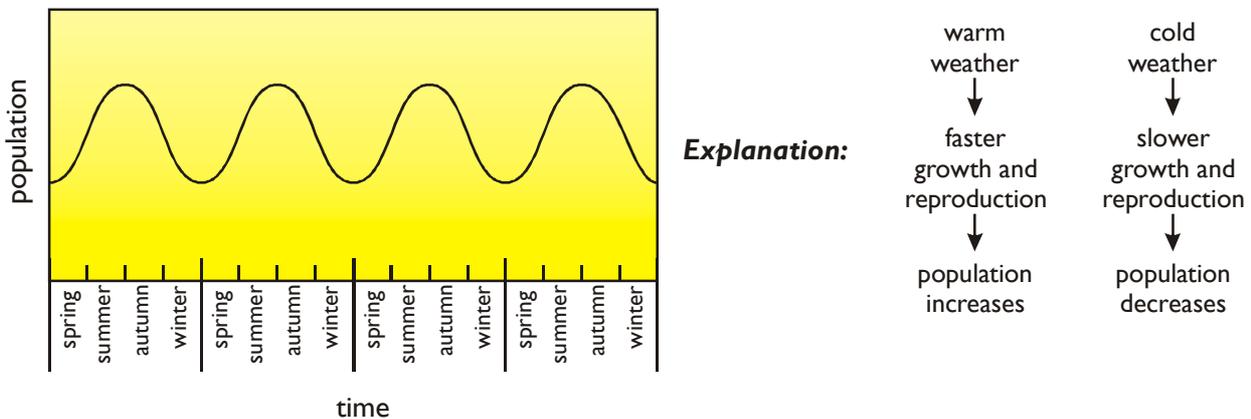
Abiotic factors

Abiotic factors are all the physical or non-living aspects to an ecosystem. These include:

- Climatic factors, such as temperature; water/humidity; light/shade; current (wind/water), frost
- Edaphic (soil) factors, such as pH; mineral supply; soil texture; soil moisture
- Topographic factors, such as altitude, slope, aspect
- Human factors, such as pollution.
- Catastrophes, such as floods and fire

Abiotic factors can vary within a habitat, giving microclimates in microhabitats, e.g. the abiotic factors under a stone are very different from those on top of an adjacent stone wall. Abiotic factors tend to be density-independent factors, i.e. the size of the effect is independent of the size of the population. For example a low light intensity will limit plant growth regardless of the number of plants present.

Many abiotic factors vary with the seasons, and this can cause a periodic oscillation in the population size.



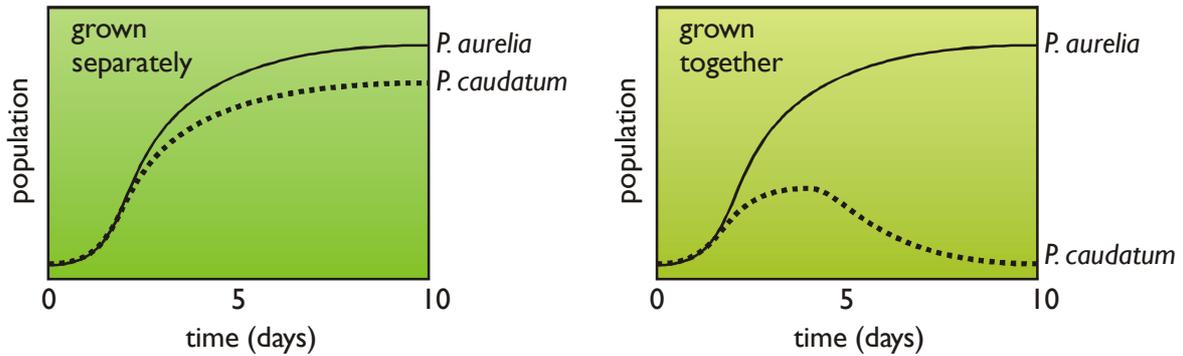
This is only seen in species with a short life cycle compared to the seasons, such as insects. Species with long life cycles (longer than a year) do not change with the seasons like this.

Biotic factors

Biotic factors are all the living aspects of an ecosystem, i.e. food, competitors, predators, parasites and pathogens. Biotic factors tend to be density-dependent factors, i.e. the size of the effect depends on the size of the population. For example competition will be greater the greater the population.

Interspecific Competition

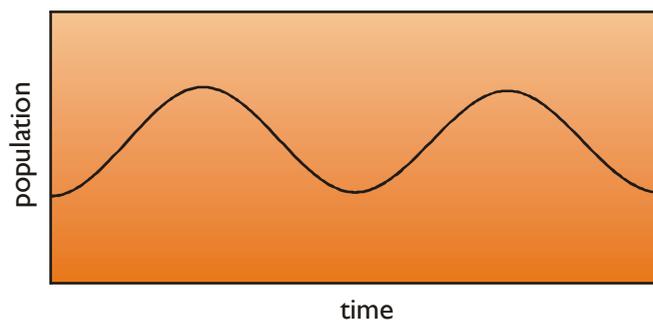
Interspecific competition is competition for resources (such as food, space, water, light, etc.) between members of different species, and in general one species will out-compete another one. Interspecific competition tends to have a dramatic effect on populations. This can be demonstrated in the field or in a controlled laboratory habitat, using flasks of the protozoan *Paramecium*, which eats bacteria. Two different species of *Paramecium* grow well in lab flasks when grown separately, but when grown together *P.aurelia* out-competes *P.caudatum* for food, so the population of *P.caudatum* falls due to interspecific competition:



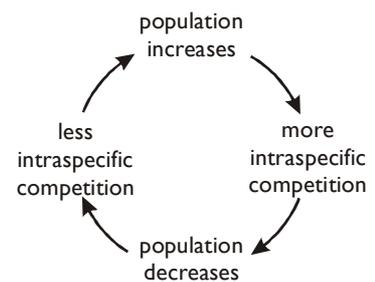
Intraspecific Competition

Intraspecific competition is competition for resources between members of the same species. This is more significant than interspecific competition, since members of the same species have the same niche and so compete for exactly the same resources.

Intraspecific competition tends to have a stabilising influence on population size because it is density-dependent. If the population gets too big, intraspecific competition increases, so the population falls again. If the population gets too small, intraspecific competition decreases, so the population increases again:



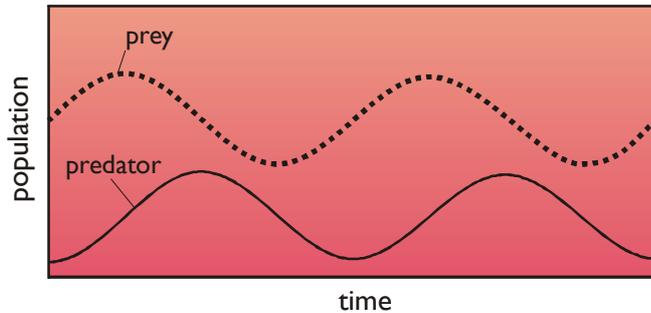
Explanation:



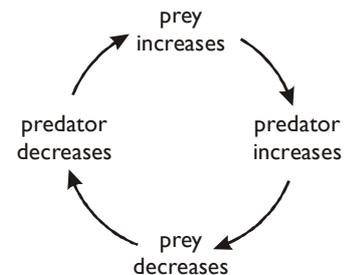
Intraspecific competition is also the driving force behind natural selection, since the individuals with the “best” genes are more likely to win the competition and pass on their genes. Some species use aggressive behaviour to minimise real competition. Ritual fights, displays, threat postures are used to allow some individuals (the “best”) to reproduce and exclude others (the “weakest”). This avoids real fights or shortages, and results in an optimum size for a population.

Predation

The populations of predators and their prey depend on each other, so they tend to show cyclical changes. This has been famously measured for populations of lynx (predator) and hare (prey) in Canada, and can also be demonstrated in a lab experiment using two species of mite: *Eotetranchus* (a herbivore) and *Typhlodromus* (a predator). If the population of the prey increases, the predator will have more food, so its population will start to increase. This means that more prey will be eaten, so its population will decrease, so causing a cycle in both populations:

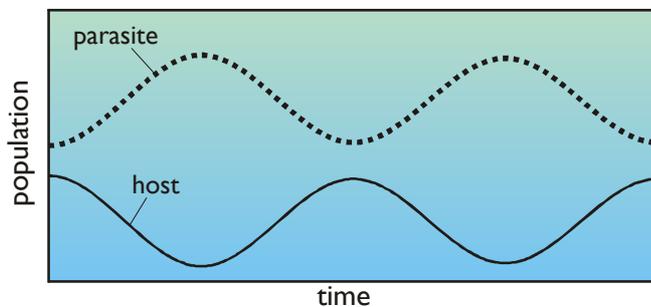


Explanation:

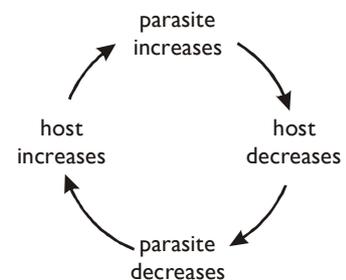


Parasitism and Disease

Parasites feed on larger host organisms, harming them. Parasites and their hosts have a close symbiotic relationship, so their populations also oscillate. This is demonstrated by winter moth caterpillars (the host species) and wasp larvae (parasites on the caterpillars). If the population of parasite increases, they kill their hosts, so their population decreases. This means there are fewer hosts for the parasite, so their population decreases. This allows the host population to recover, so the parasite population also recovers:



Explanation:



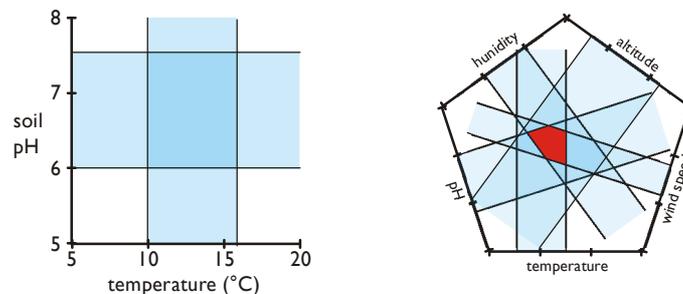
A similar pattern is seen for pathogens and their hosts.

In harsh environments (very cold, very hot, very dry, very acid, etc.) only a few species will have successfully adapted to the conditions so they will not have much competition from other species, but in mild environments lots of different species could live there, so there will be competition. In other words in harsh environments abiotic factors govern who survives, while in mild environments biotic factors (such as competition) govern who survives.

The Ecological Niche

An organism's niche refers to the biotic and abiotic factors that the organism needs in its habitat. It would be impossible to have a complete list of all the required factors, so we tend to focus on a few aspects that interest us. We often focus on an organism's role in its food chain (e.g. producer, predator, parasite, etc.) and might include more details like the specific food (e.g. leaf-eater, insectivore, grassland grazer etc.). Alternatively we might be interested in details of an organism's reproduction method, or hunting strategy, or geographic location, or seasonal distribution, or migration pattern, or host specificity, or microhabitat, etc.

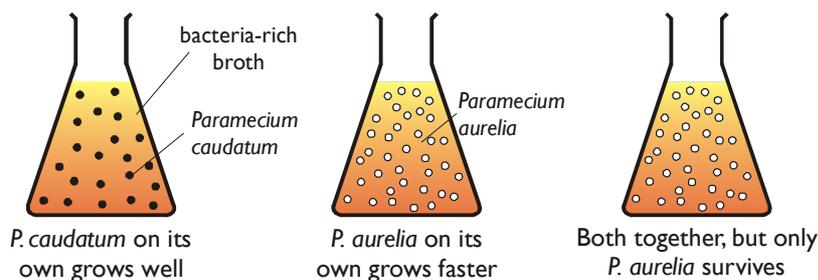
The abiotic factors that comprise an organism's niche can be shown on a graph. For example, if a particular plant can only grow in a temperature range of 10–17°C and a soil pH of 6–7.5, then these ranges can be plotted on two axes of a graph, and where they intersect (the shaded box in the graph on the left) shows those aspects of the plant's niche. We can add further axes to show the suitable ranges of other factors like humidity, light intensity and altitude, and so get a more detailed description of the niche (graph on right).



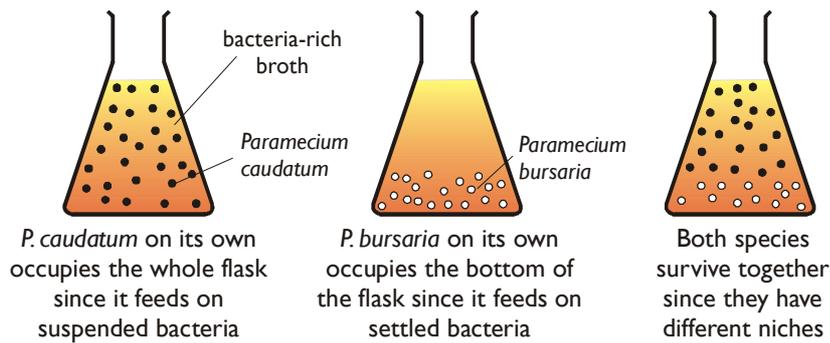
Members of the same population (i.e. same species) always have the same niche, so the niche of a population is genetically-determined, not learned. Successful organisms are always well-adapted to their niche, so a niche can also be thought of as all the biotic and abiotic factors to which members of a population are adapted.

Identifying the different niches in an ecosystem helps us to understand the interactions between populations. The niche concept was investigated in some classic experiments in the 1930s by Gause. He used flasks of different species of the protozoan *Paramecium*, which eats bacteria.

Experiment. I:



Conclusion: These two species of *Paramecium* share the same niche, so they compete. *P. aurelia* is faster-growing, so it out-competes *P. caudatum*.

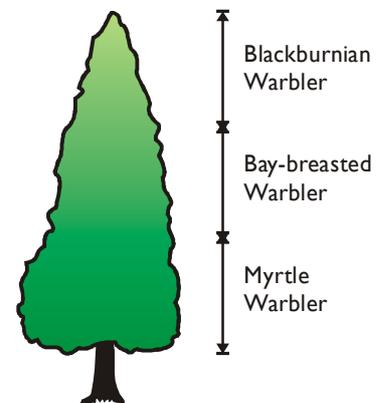
Experiment. 2:

Conclusion: These two species of *Paramecium* have slightly different niches, so they don't compete and can coexist.

It is important to understand the distribution in experiment 2. *P. caudatum* lives in the upper part of the flask because only it is adapted to that niche and it has no competition. In the lower part of the flask both species could survive, but only *P. bursaria* is found because it out-competes *P. caudatum*. If *P. caudatum* was faster-growing it would be found throughout the flask.

The niche concept is summarised in the competitive exclusion principle: **Two species cannot coexist in the same habitat if they have the same niche.** They will compete, and one species will win the competition. This principle also works in reverse: if two species are observed to compete then they must have the same niche.

- Species with narrow niches are called specialists. Many different specialists can coexist in the same habitat because they are not competing, so this can lead to high diversity. For example warblers in a coniferous forest feed on insects found at different heights (see right). Specialists rely on a constant supply of their food, so are generally found in abundant, stable habitats such as the tropics.
- Species with broad niches are called generalists. Generalists in the same habitat will compete, so there can only be a few, so this can lead to low diversity. Generalists can cope with a changing food supply (such as seasonal changes) since they can switch from one food to another or even one habitat to another (for example by migrating).



The competitive exclusion principle may apply whenever a new species is introduced to an ecosystem. For example American grey squirrels are out-competing and excluding the native red squirrels in England, and the Australian barnacle is out-competing and excluding the native English species on rocky shores. These native species are declining and may eventually become extinct.

Ecological Succession

Ecosystems are not fixed, but constantly change with time. This change is called succession. Different species of plants naturally colonise a habitat in a predictable order, until finally a stable community is reached, called the climax community. Each plant species in turn changes its environment (e.g. by creating deeper soil, or providing shade), making the environment more suitable for new species to colonise. These new species are usually bigger plants with a larger photosynthetic area, so they outcompete and replace the older species. So each plant effectively causes its own demise. The plants colonising early in succession (the pioneer species) tend to be small and fast growing, with shallow roots and wind-dispersed seeds. The plants colonising late in succession tend to be tall and slow growing, with deep roots and animal-dispersed seeds.

The successive stages are called seral stages, or seral communities, and the whole succession is called a sere. It will usually take a few hundred years to reach a stable climax community. The climax community is usually a forest, though this varies depending on the climate and the underlying rock. In England the natural climax community is oak, ash or beech woodland, and in the highlands of Scotland it is pine forests.

As the succession proceeds the habitat becomes less harsh and the abiotic factors less hostile. For example daily temperature fluctuations decrease (due to shade); water is more easily available (since it is retained in soil) and nitrates increase (due to nitrogen fixation and decay). These changes are what allow more plant species to colonise. As the plant community becomes more diverse, the animal community also becomes more diverse, since there is a greater variety of food for primary consumers and therefore a greater variety of food for secondary consumers. There is also a greater diversity of niches in the more complex ecosystem. The climax community supports a complex food web, which also aids stability.

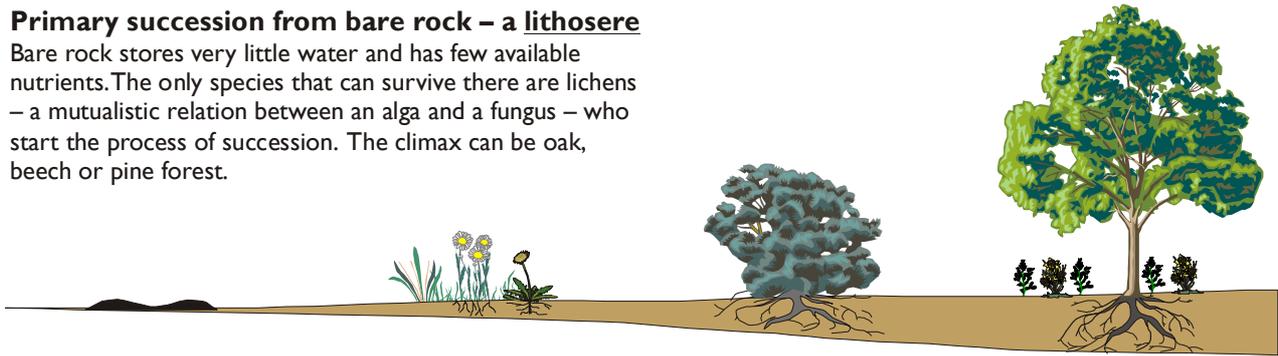
There are two kinds of succession:

- Primary succession starts with bare rock or sand, such as behind a retreating glacier, after a volcanic eruption, following the silting of a lake or seashore, on a new sand dune, or on rock scree from erosion and weathering of a mountain.
- Secondary succession starts with soil, but no (or only a few) species, such as in a forest clearing, following a forest fire, or when soil is deposited by a meandering river.

Examples of succession

Primary succession from bare rock – a lithosere

Bare rock stores very little water and has few available nutrients. The only species that can survive there are lichens – a mutualistic relation between an alga and a fungus – who start the process of succession. The climax can be oak, beech or pine forest.



1

lichens and mosses

The first pioneers are lichens, who can absorb the scarce water from the bare rock. Mosses can then grow on top of the lichens. These species are very small, slow-growing, wind-dispersed and tolerant of extreme conditions. They start to weather the rock by secreting acids, and so begin to form a very thin soil.

2

grasses and herbs

The next colonisers are grasses and ferns, followed by small herbaceous plants such as dandelion and nettles. These species have a larger leaf area, so they grow fast and out-compete the pioneers. Their larger roots weather the rock and add more detritus, adding inorganic and organic matter to the soil, which now holds more water.

3

shrubs and bushes

Larger plants (shrubs) such as bramble, gorse, hawthorn, broom and rhododendron can now grow in the thicker soil. These species have larger, animal-dispersed seeds and they grow faster and taller, out-competing the smaller herbs.

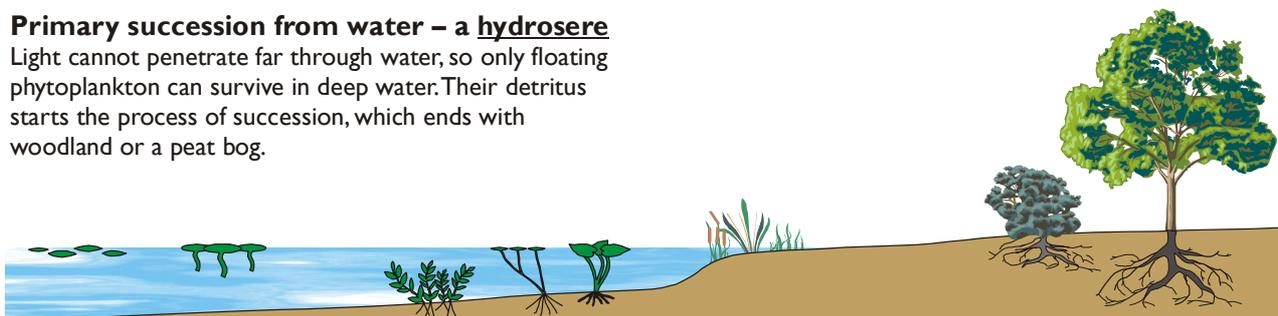
4

woodland

Trees grow slowly, but eventually shade and out-compete the shrubs, which are replaced by shade-tolerant forest-floor species. A complex layered community is now established with many trophic levels and interactions. This is the climax community.

Primary succession from water – a hydrosere

Light cannot penetrate far through water, so only floating phytoplankton can survive in deep water. Their detritus starts the process of succession, which ends with woodland or a peat bog.



1

plankton

Floating phytoplankton colonise deep water using wind-dispersed spores. When they die they sink to the bottom, forming humus, which combines with silt deposited by rivers to form mud that builds up on the bottom.

2

rooted aquatic plants

As the mud builds up, the water becomes shallower, allowing rooted plants to grow. These include submerged species, like pondweed, and species with floating leaves, like lilies. Their root systems trap more silt and their faster growth results in more detritus settling to the bottom.

3

swamp and marsh

Eventually the sediment rises out of the water to form a waterlogged soil. Reed grasses and sedges colonise to form a reed marsh. Their roots bind the mud together to form semisolid soil, and the increased rate of transpiration starts to dry the soil.

4

woodland

As the soil dries it can be colonised by more terrestrial species. First herbs replace the marsh vegetation then shrubs replace the herbs and eventually trees replace the shrubs.

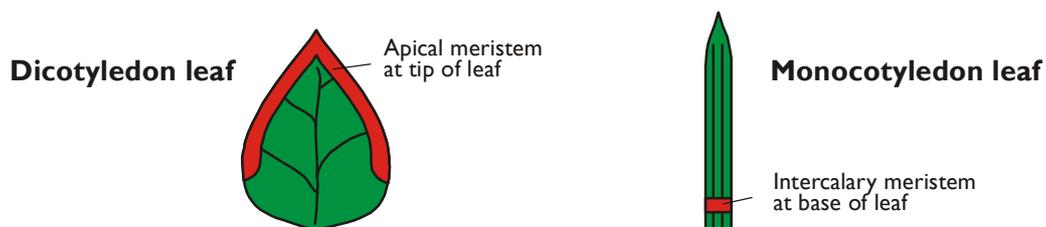
Human effects on succession

At the end of the last ice age twelve thousand years ago, Britain was a lifeless rock, scoured bare by the retreating glaciers. Over the following centuries succession led to the climax forest communities that early human settlers found and settled. In Roman times the country was covered in oak and beech woodlands with herbivores such as deer, omnivores such as bear and carnivores such as wolves and lynxes. It was said that a squirrel could travel from coast to coast without touching ground.

Humans interfere with succession, and have done so since the development of farming, by cutting down forests to make farmland. This deforestation has continued until today there are few examples of a natural climax left in the UK, except perhaps small areas of the Caledonian pine forest in the Scottish Highlands. All landscapes today like woodland, grassland, moorland, farmland and gardens are all maintained at pre-climax stages by constant human interventions, including ploughing, weeding, herbicides, burning, mowing, crop planting, grazing animals and dredging waterways. These interventions cause a deflected succession, resulting in a plagioclimax.

Why does grazing stop succession at the grassland stage?

- Herbs, shrubs, trees and the later species of plants are mostly dicotyledons (broad-leaved plants), which have strong, vertical stems and grow from apical meristems at the tips of their shoots and leaves. Grazing animals eat these apical meristems or uproot the whole plant, so the plants die.
- Grasses on the other hand are monocotyledons (narrow-leaved plants), which have horizontal stems and grow from intercalary meristems at the base of their shoots and leaves. Grazers cannot eat these intercalary meristems or uproot the plants, so grasses can continue to grow.



Conservation

Conservation is the management of our environment to maintain biodiversity. Recall from unit 2 that biodiversity encompasses genetic diversity (the variety of alleles within a species), species diversity (the variety of species within a habitat) and habitat diversity (the variety of habitats within an ecosystem).

It is important to conserve all three aspects. The global gene pool is a resource for learning more about life on Earth, and some genes may be able to provide us with useful products for medicine and biotechnology. To maintain the gene pool we need to preserve species diversity and to conserve species diversity we must provide suitable niches for all species by preserving habitat diversity. So a key aim of conservation is to prevent further destruction of habitats and preserve as wide a range of habitats as possible.

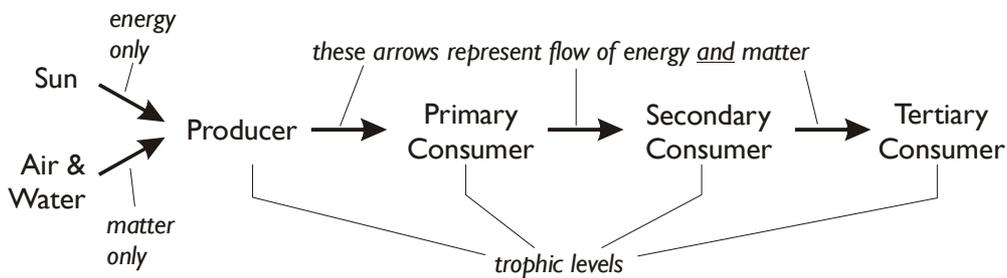
Conservation is therefore not a matter of leaving the environment untouched, which would result in a small range of climax communities. Instead conservation involves active intervention to manage succession and maintain a wide variety of different plagioclimaxes. These man-made habitats have been found to be useful to humans for hundreds or thousands of years, while still supporting a wide range of organisms. So it is often possible to keep the land as a productive resource, but in a sustainable way that maintains biodiversity. For example:

- Moorland is maintained by periodic burning. Fire kills tree saplings but not heather, which is fire-resistant and re-grows after a few weeks.
- Grassland is maintained by grazing animals, which prevent the growth of shrubs and trees, but allows grasses to grow. Where succession has been allowed to take place, grassland can be restored by felling and removing the shrubs and trees. Grazing by sheep and rabbits leaves grass particularly short and creates unique environments, such as the chalk grasslands in the North and South Downs in southern England. These chalk grasslands are some of the most biodiverse environments in the UK, supporting 30-40 species m⁻².
- Wetlands are maintained by dredging to prevent silting up and succession, and by ensuring the water supply is free from pollution by farms and factories upstream.
- Woodland is maintained by replacing non-native conifer plantations with native broad-leaved trees and reducing density by thinning. Thinning allows more light to reach the ground layer, encouraging the growth of shrubs and wildflowers. A forest can be managed by coppicing and pollarding, which allow timber to be harvested, while conserving the forest.
- Hedgerows and field margins are small but important habitats for conserving diversity in farmland. They are maintained by occasional cutting back to prevent succession to a climax forest.

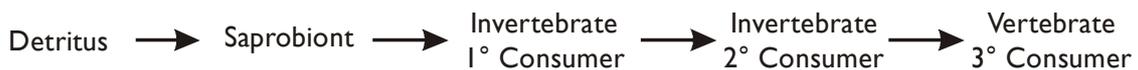
Food Chains

The many relationships between the members of a community in an ecosystem can be described by food chains and webs. Each stage in a food chain is called a trophic level, and the arrows represent the flow of energy and matter through the food chain.

Food chains always start with photosynthetic producers (plants, algae, plankton and photosynthetic bacteria) because, uniquely, producers are able to extract both energy and matter from the abiotic environment (energy from the sun, and 98% of their dry mass from carbon dioxide in the air, with the remaining 2% from water and minerals in soil). All other living organisms get both their energy and matter by eating other organisms. All living organisms need energy and matter from their environment. Matter is needed to make new cells (growth) and to create new organisms (reproduction), while energy is needed to drive all the chemical and physical processes of life, such as biosynthesis, active transport and movement.



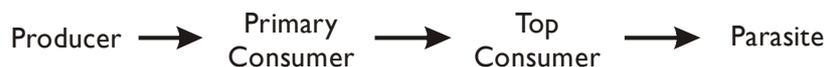
This represents a “typical” terrestrial food chain, but there are many other possible food chains. In particular, detritus (such as dead leaves and animal waste) plays an important role in aquatic food chains, with small invertebrate consumers feeding on microbial decomposers (saprobionts).



Deep water ecosystems have few producers (since there is little light), so the main food source for consumers is detritus washed down from rivers.



The top of a food chain is often not a top consumer, but rather scavengers or parasites feeding on them.



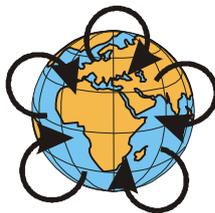
Matter and Energy

Organisms need both matter and energy from their environment, and, before we look at matter and energy transfer in more detail, it is important to be clear about the difference between the two. Matter and energy are quite different things and cannot be inter-converted.

Matter

Matter (chemicals) is measured in kilograms and comes in three different states (solid, liquid and gas). It cannot be created, destroyed or used up. The Earth is a closed system with respect to matter, in other words the total amount of matter on the Earth is constant. The matter of a living organism is called its biomass. Matter (and especially the biochemicals found in living organisms) can contain stored chemical energy, so a cow contains biomass (matter) as well as chemical energy stored in its biomass.

Matter Cycles

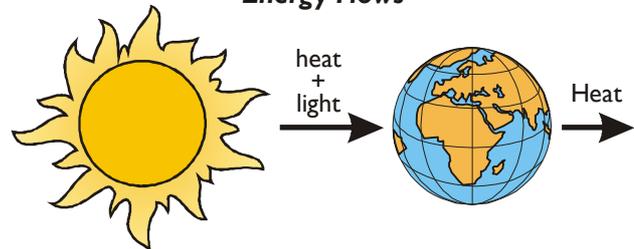


Matter cycles between living and non-living things. But no new matter reaches the Earth, and none leaves.

Energy

Energy is measured in joules and comes in many different forms (such as heat, light, chemical, potential, kinetic, etc.). These forms can be inter-converted, but energy can never be created, destroyed or used up. If we talk about energy being “lost”, we usually mean as heat, which is radiated out into space. The Earth is an open system with respect to energy, in other words energy can enter and leave the Earth. Energy is constantly arriving on Earth from the sun, and is constantly leaving the earth as heat, but the total amount of energy on the earth remains roughly constant.

Energy Flows



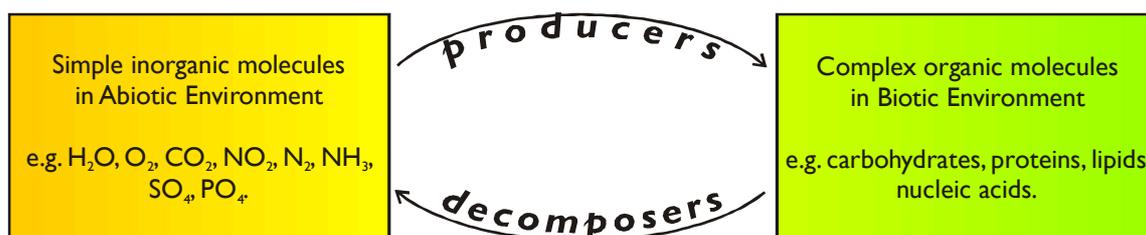
Energy is constantly arriving from the sun, passing through living organisms, and leaving the Earth as heat.

There are many ecological terms relating to food chains, and most describe aspects of an organism's niche:

Producer	An organism that produces food from carbon dioxide and water using photosynthesis. Can be plant, algae, plankton or bacteria (a.k.a. primary producer).
Consumer	An animal that eats other organisms
Herbivore	A consumer that eats plants (= primary consumer).
Carnivore	A consumer that eats other animals (= secondary consumer).
Top carnivore	A consumer at the top of a food chain with no predators.
Omnivore	A consumer that eats plants or animals.
Vegetarian	A human that chooses not to eat animals (humans are omnivores)
Autotroph	An organism that manufactures its own food (= producer)
Heterotroph	An organism that obtains its energy and mass from other organisms (=consumers + decomposers)
Flora	old-fashioned/literary term for plants
Fauna	old-fashioned/literary term for animals
Plankton	Microscopic marine organisms.
Phytoplankton	"Plant plankton" i.e. microscopic marine producers.
Zooplankton	"Animal plankton" i.e. microscopic marine consumers.
Predator	An animal that hunts and kills animals for food.
Prey	An animal that is hunted and killed for food.
Scavenger	An animal that eats dead animals, but doesn't kill them
Detritus	Dead and waste matter that is not eaten by consumers
Carrion	Alternative word for detritus
Decomposer	An organism that uses detritus for nutrition (= detritivores + saprobionts)
Detritivore	An animal that eats detritus.
Saprobiont	A microbe (bacterium or fungus) that lives on detritus (a.k.a. saprotroph)
Symbiosis	Organisms living together in a close relationship (=mutualism, commensalism, parasitism, pathogen).
Mutualism	Two organisms living together for mutual benefit.
Commensalism	Relationship in which only one organism benefits
Parasite	An organism that feeds on a larger living host organism, harming it
Pathogen	A microbe that causes a disease.

Nutrient Cycles in Ecosystems

Matter cycles between the biotic environment and in the abiotic environment. Simple inorganic molecules (such as CO_2 , N_2 and H_2O) are assimilated (or fixed) from the abiotic environment by producers and microbes, and built into complex organic molecules (such as carbohydrates, proteins and lipids). (In science organic compounds contain carbon-carbon bonds, while inorganic compounds don't.) These organic molecules are passed through food chains and eventually returned to the abiotic environment again as simple inorganic molecules by decomposers. Without either producers or decomposers there would be no nutrient cycling and no life.



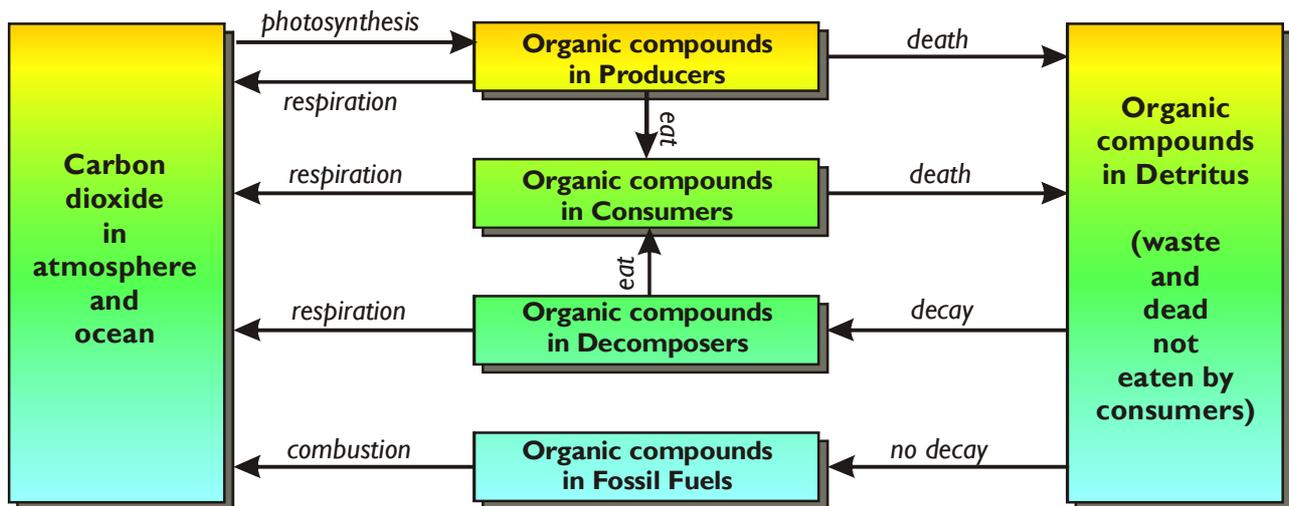
The simple inorganic molecules are often referred to as nutrients. Nutrients can be grouped into three classes:

class	elements	% of biomass
Major Nutrients	C O H	99.4%
Macro Nutrients	N S P K Ca Mg Al Si	0.5%
Micro Nutrients	Na Fe Co Cu Zn Mn Sn Va Cl F I	0.1%

The major nutrients are taken from the environment in the form of carbon dioxide (C and O) and water (H). All the other nutrients are usually required as soluble mineral ions, so are often referred to as "minerals". While the major nutrients are obviously needed in the largest amounts, the growth of producers is often limited by the availability of macro nutrients such as nitrogen and phosphorus.

Detailed nutrient cycles can be constructed for elements such as carbon, nitrogen, oxygen or sulphur, or for compounds such as water, but they all have the same basic pattern as the diagram above. We shall only study the carbon and nitrogen cycles in detail.

The Carbon Cycle



As this diagram shows, there are really many carbon cycles, with time scales ranging from minutes to millions of years.

Photosynthesis is the only route by which carbon dioxide is “fixed” into organic carbon compounds. Terrestrial producers (mainly forests) account for about 50% of all carbon fixation globally, with the other 50% due to marine microbial producers (phytoplankton). Photosynthesis is balanced by respiration, decay and combustion, which all return carbon dioxide to the atmosphere. Different ecosystems have a different balance:

- A carbon source is an ecosystem that releases more carbon as carbon dioxide than it accumulates in biomass over the long term. Carbon sources include farmland (since crops are eaten and respired quickly and decay is encouraged by tilling) and areas of deforestation (since the tree biomass is burned or decayed).
- A carbon neutral ecosystem is one where carbon fixation and carbon release are balanced over the long term. Carbon neutral ecosystems include mature forests, where new growth is balanced by death and decay.
- A carbon sink is an ecosystem that accumulates more carbon in biomass than it releases as carbon dioxide over the long term. This accumulation happens when the conditions are not suitable for decomposers (too cold, too dry, too acidic, etc). Carbon sinks include peat bogs (since the soil is too acidic for decay), the ocean floor (since it is too cold and anaerobic for detritus to decay); and growing forests (since carbon is being incorporated into growing biomass). In a carbon sink the carbon remains fixed in organic form and can even form a fossil fuel given enough time. The vast fern swamps of the carboniferous era (300MY ago) were carbon sinks and gave rise to all the coal seams we mine today. The recent mining and burning of fossil fuels has significantly altered the carbon cycle by releasing this carbon into the atmosphere again, causing a 15% increase in CO_2 in just 200 years (see p 48).

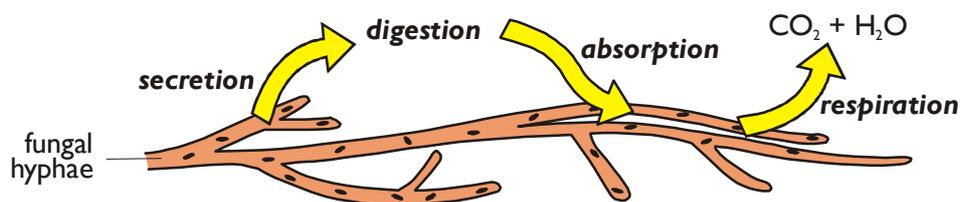
Decay

Decay (also known as decomposition, putrefaction or rotting) is the breakdown of detritus by organisms collectively called decomposers. There are two groups of decomposers: saprobionts and detritivores.

Saprobionts

Saprobionts (or saprotrophs) are microbes (fungi and bacteria) that live on detritus. Saprobionts use saprobiotic nutrition, which means they do not ingest their food, but instead use extracellular digestion, secreting digestive enzymes into the detritus that surrounds them and absorbing the soluble products. The absorbed products are then further broken down in aerobic respiration to inorganic molecules such as carbon dioxide, water and mineral ions. Only a few bacteria possess the cellulase enzymes required to break down the plant fibres that comprise much of the detritus biomass. Herbivorous animals such as cows and termites depend on these bacteria in their guts.

In aquatic ecosystems the main saprobionts are bacteria, while in terrestrial ecosystems the main saprobionts are fungi. Fungi are usually composed of long thin threads called hyphae. These hyphae grow quickly throughout soil giving fungi a large surface area to volume ratio. The total amount of fungi in the environment is surprising: there is a similar mass of fungal biomass growing underground beneath a forest than there is plant biomass growing above ground (see also mycorrhizae on p 31).



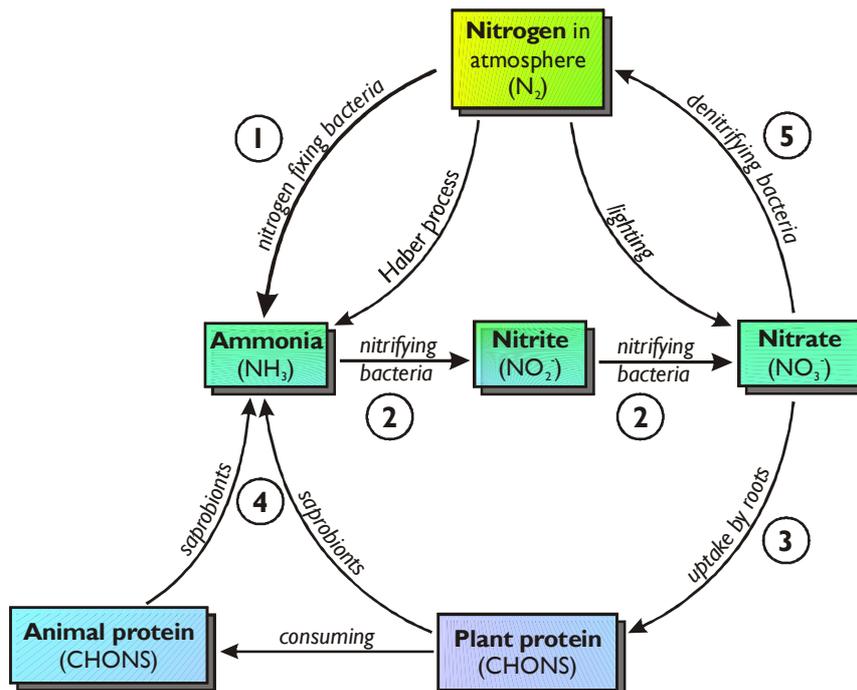
Detritivores

Detritivores are small invertebrate animals (such as earthworms and woodlice) that eat detritus. Like all animals, they use holozoic nutrition, i.e. they ingest food, digest it in a gut, absorb the soluble products and egest the insoluble waste. This egesta consists largely of plant fibres (cellulose and lignin), which animals can't digest. Detritivores speed up decomposition by helping saprobionts:

- Detritivores physically break up large plant tissue (like leaves or twigs) into much smaller pieces, which they egest as faeces. The faeces has a larger surface area making it more accessible to the saprobionts.
- Detritivores aerate the soil, which helps the saprobionts to respire aerobically.
- Detritivores excrete useful minerals such as urea, which saprobionts can metabolise.

Neither saprobionts nor detritivores can control their body temperature, so their activity (metabolism and reproduction) depends on the environmental temperature. Decay therefore happens much more rapidly in summer than in winter.

The Nitrogen Cycle



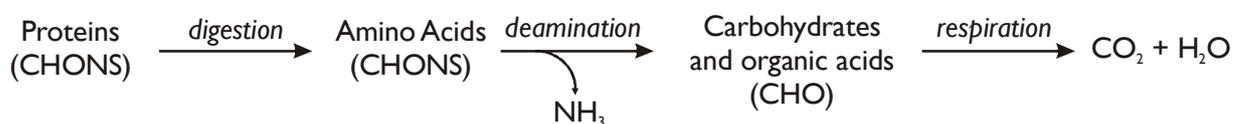
Nitrogen is needed by all living organisms to make proteins (CHONS) and nucleic acids (CHONP). There are several different forms of inorganic nitrogen that occur in the nitrogen cycle: N₂, NO₂ and NH₃. The word "nitrogen" is confusingly used to mean nitrogen atoms (N) or nitrogen molecules (N₂). So for example proteins contain nitrogen atoms but not nitrogen molecules. To avoid confusion, always refer to N₂ as nitrogen gas or N₂.

1. **Nitrogen Fixation.** 78% of the atmosphere is nitrogen gas (N₂), but the triple bond linking the two nitrogen atoms makes it a very stable molecule, which doesn't readily take part in chemical reactions. N₂ therefore can't be used by plants or animals as a source of nitrogen. The nitrogen in N₂ is "fixed" into useful compounds by nitrogen fixing bacteria. They reduce nitrogen gas to ammonia (N₂ + 6H → 2NH₃), which dissolves to form ammonium ions (NH₄⁺). This reaction is catalysed by the enzyme nitrogenase and it requires a great deal of energy: 15 ATP molecules need to be hydrolysed to fix each molecule of N₂.

Some nitrogen-fixing bacteria are free-living in soil, but most live in colonies inside the cells of root nodules of leguminous plants such as clover or peas. This is a classic example of mutualism, where both species benefit. The pea plants gain a source of useful nitrogen from the bacteria, while the bacteria gain carbohydrates from the plant, which they respire to make the large amounts of ATP they need to fix nitrogen.

Nitrogen gas can also be fixed to ammonia by humans using the Haber process ($N_2 + 3H_2 \rightarrow 2NH_3$) to make nitrogenous fertilisers, which are spread on to soil. Today almost a third of all nitrogen fixed is fixed by the Haber process. The long-term effects of this increase in nitrogen fixing remain to be seen. Nitrogen can also be fixed by oxidising it to nitrate ($N_2 + 2O_2 \rightarrow 2NO_2$). This reaction happens naturally by lightning and was probably very important in the early earth's atmosphere, but is not a significant process now.

2. **Nitrification.** Nitrifying bacteria can oxidise ammonia to nitrate in two stages: first forming nitrite ions ($NH_4^+ \rightarrow NO_2^-$) then forming nitrate ions ($NO_2^- \rightarrow NO_3^-$). This oxidation reaction is exothermic, releasing energy, which these bacteria use to make ATP, instead of using respiration.
3. **Assimilation.** Plants are extremely self-sufficient: they can make carbohydrates and lipids from CO_2 and H_2O , but to make proteins and nucleotides they need a source of nitrogen. Plants require nitrogen in the form of dissolved nitrates, and the supply of nitrates is often so poor that it limits growth (which is why farmers add nitrate fertilisers to crops). Plants use active transport to accumulate nitrate ions in their root hair cells against a concentration gradient. Most plant species have symbiotic fungi associated with their roots called mycorrhizae. These mycorrhizae aid mineral absorption since the hyphae are thinner than roots and so have a larger surface area : volume ratio. Some plants living in very poor soils have developed an unusual strategy to acquire nitrogen: they trap and digest insects. These so-called carnivorous plants don't use the insects as a main source of nutrition as a consumer would do, but just as a source of nitrogen-containing compounds.
4. **Ammonification.** Microbial saprobionts break down proteins in detritus to form ammonia in two stages: first they digest proteins to amino acids using extracellular protease enzymes, and then they remove the amino groups from amino acids using deaminase enzymes to form ammonia (NH_4^+). The deaminated amino acids, now containing just the elements CHO, are respired by the saprobionts to CO_2 and H_2O (see the carbon cycle).

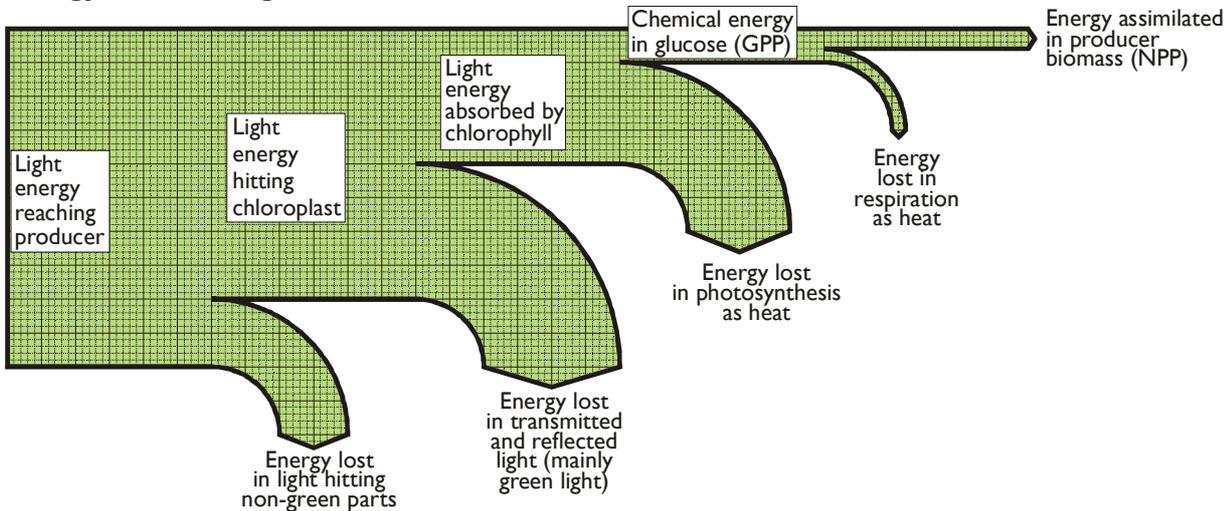


5. **Denitrification.** The anaerobic denitrifying bacteria convert nitrate to N_2 and NO_x gases, which are then lost to the air. This represents a constant loss of "useful" nitrogen from soil, and explains why nitrogen fixation by the nitrifying bacteria and fertilisers are so important.

Energy Flow in Ecosystems

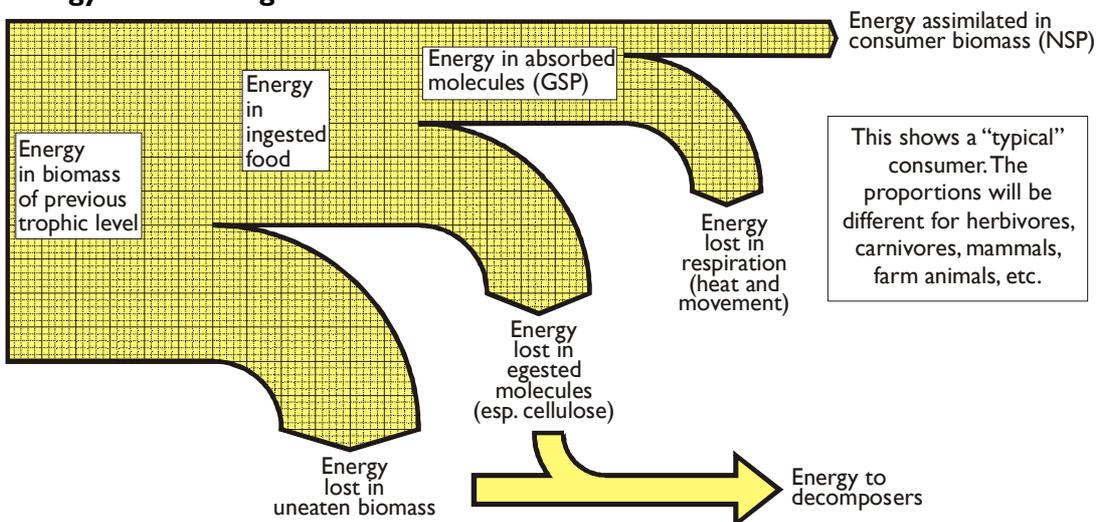
The flow of energy through a trophic level in a food chain can be shown in detail using Sankey diagrams. These show the possible fates of energy as arrows, with the width of the arrow representing the amount of energy.

Energy Flow through a Producer



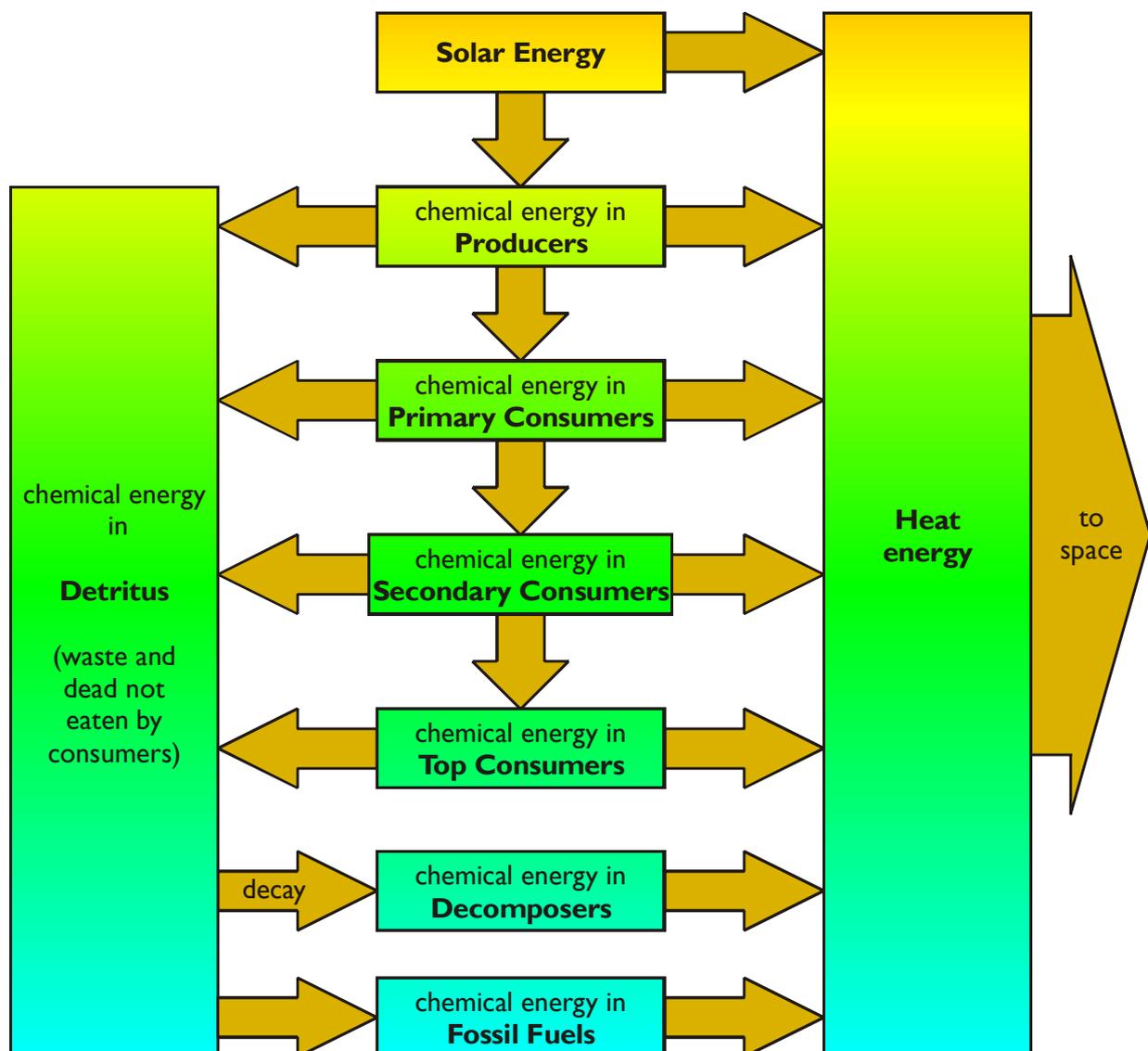
Energy enters food chains in the form of light energy. Three things can happen to light when it reaches an object: it can be reflected, transmitted or absorbed. Only light energy that is absorbed by chlorophyll molecules in producers can be converted into chemical energy in glucose and so enter the food chain. Light energy that is not absorbed by chlorophyll will eventually be absorbed by other objects on the ground, such as water, rocks or animals, and will be converted to heat. A lot of this energy is used to evaporate water and so drive the water cycle. More energy is lost as heat during photosynthesis and respiration, so around 99% of the light energy reaching the Earth is converted to heat, with less than 1% being “fixed” in producer biomass.

Energy Flow through a Consumer



Consumers have an easier job, since they take in concentrated chemical energy in the form of the organic molecules that make up the biomass of the plants or animals they eat. As we've already seen a lot of biomass is not absorbed by consumers (plant fibre, wood, bone, fur, etc.) and the energy in this biomass is passed on to decomposers, who can use it. And much of the energy that is absorbed is lost as heat through the various metabolic reactions, especially respiration and friction due to movement. The heat energy is given out to the surroundings by radiation, convection and conduction, and cannot be regained by living organisms. These losses are particularly big in warm-blooded and very active animals. Only 1-5% of the available energy is assimilated into consumer biomass, which can then be consumed by the next animal in the food chain.

This diagram shows the energy flow through a whole food chain. Eventually all the energy that enters the ecosystem will be converted to heat, which is lost to space.

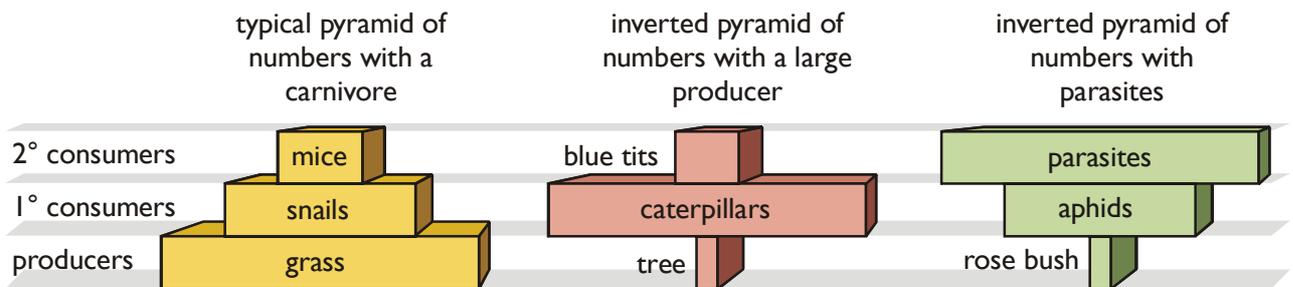


Ecological Pyramids

The transfer of energy and matter through food chains can also be displayed in ecological pyramids. There are three kinds:

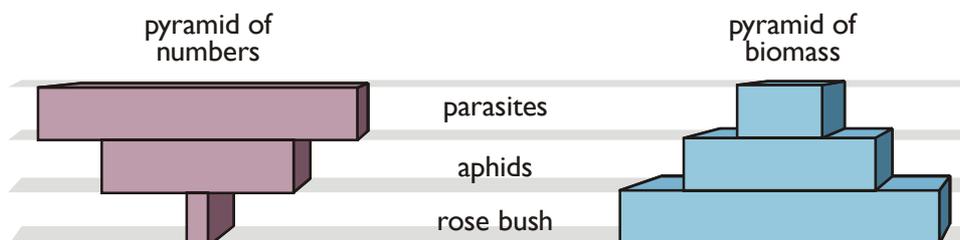
1. Pyramids of Numbers.

These show the numbers of organisms at each trophic level in a food chain. The widths of the bars represent the numbers using a linear or logarithmic scale, or the bars may be purely qualitative. The numbers should be normalised for a given area for a terrestrial habitat (so the units would be numbers m^{-2}), or volume for a marine habitat (so the units would be numbers m^{-3}). In general as you go up a food chain the size of the individuals increases and the number of individuals decreases. So pyramids of numbers are often triangular (or pyramid) shaped, but can be almost any shape, depending of the size of the organisms. Many terrestrial producers are very large (such as trees) and many primary consumers are very small (such as insects and other invertebrates) so these differences cause inverted pyramids.



2. Pyramids of Biomass

These convey more information, since they consider the total mass of living organisms (i.e. the biomass) at each trophic level. The biomass should be dry mass (since water stores no energy) and is measured in $kg\ m^{-2}$. The biomass may be found by drying and weighing the organisms at each trophic level, or by counting them and multiplying by an average individual mass. Pyramids of biomass are usually pyramid shaped (even if the pyramid of numbers isn't), since if a trophic level gains all its mass from the level below, then it cannot have more mass than that level (you cannot weigh more than you eat).

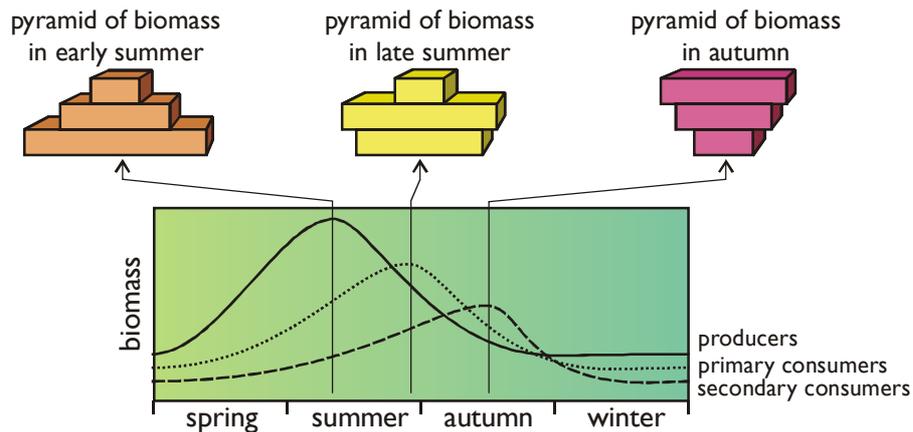


Typically, only around 10% of the biomass in each level is passed on to the next level. Mass is lost at each stage of a food chain for two reasons:

- Some of the biomass absorbed by a consumer is used in respiration and is converted to carbon dioxide and water, which are excreted (when organisms respire they lose mass!).

- Some of the biomass is simply not eaten by the consumers in the next trophic level, or is ingested but then egested again without being absorbed. This unused biomass can include plant cellulose cell walls, wood, bones, teeth, skin and hair. Many consumers are surprisingly fussy about what they eat. This biomass becomes detritus and is used by decomposers.

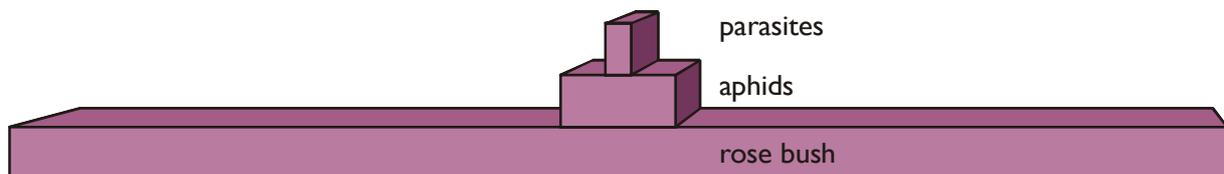
Occasionally a pyramid of biomass can be inverted. This can happen with aquatic ecosystems when growth is rapid and seasonal, so the shape of the pyramid of biomass depends on the season when the measurements were taken. For example:



If the average biomass over a whole year was measured, the pyramid biomass would be a normal shape.

3. Pyramids of Energy

These pyramids represent the flow of energy into each trophic level, so describe a period of time (usually a year). The units are usually something like $\text{kJ m}^{-2} \text{y}^{-1}$. Pyramids of energy are always pyramidal (energy can be lost but cannot be created), and are always very shallow, since the transfer of energy from one trophic level to the next is very inefficient. Typically, only around 1% of the energy in each level is passed on to the next level. The “missing” energy, which is not passed on to the next level, is lost eventually as heat.



Summary

In food chains matter is lost as:

- Carbon dioxide due to respiration
- Uneaten parts, e.g. skin, bones, teeth, shells, wood, bark.
- Waste, e.g. faeces, urine

In food chains energy is lost as:

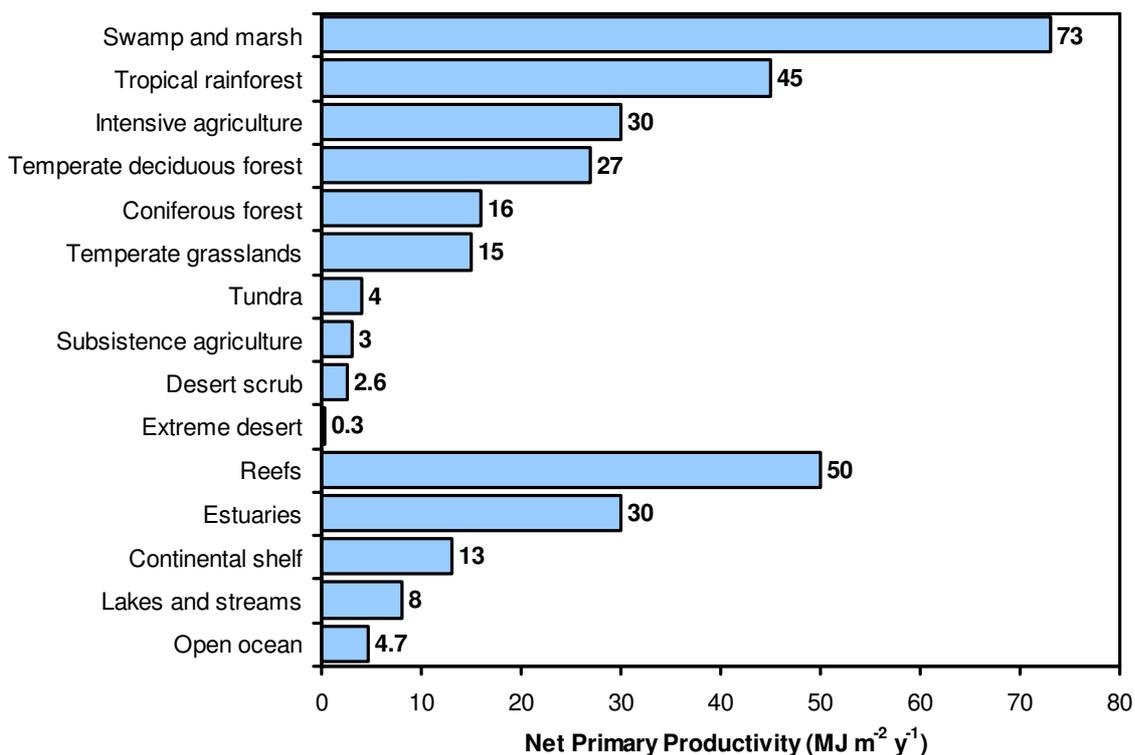
- Chemical energy in the uneaten parts.
- Movement energy of consumers.
- Heat energy, especially in homeothermic animals

Productivity

The productivity (or production) of an ecosystem is the amount of biomass produced by that ecosystem each year. Productivity can be measured in units of biomass (e.g. $\text{kg m}^{-2}\text{y}^{-1}$), or units of energy (e.g. $\text{MJ m}^{-2}\text{y}^{-1}$). We can measure the amount of plant biomass produced (primary productivity), or the amount of consumer biomass produced (secondary productivity). The amount of energy fixed by producers in photosynthesis and stored as chemical energy in glucose is called the gross primary productivity (GPP), while the amount of energy absorbed by consumers is called the gross secondary productivity (GSP). But, as the Sankey diagrams showed, some of this gross productivity is lost as heat (via respiration) and so is not available to the next level in the food chain. The amount of energy actually accumulated in producer or consumer biomass, and available to be passed on to the next trophic level, is called the net primary productivity (NPP) or net secondary productivity (NSP) respectively. These terms are shown in the two Sankey diagrams above. Gross and net productivity are related by this equation:

$$\text{Net productivity} = \text{Gross productivity} - \text{Losses due to respiration and heat}$$

NPP tells us how good an ecosystem is at fixing solar energy, so it can be used to compare the efficiency of different ecosystems. This chart shows the NPP of a variety of ecosystems:



As you would expect deserts, with very few producers, have very low productivity; while tropical rainforests, with several layers of producers and complex food webs, have a very high productivity. In fact tropical rainforests contribute about 26% of the Earth's total productivity even though they cover only 5% of the Earth's surface.

Intensive Farming

Productivity is of particular interest to farmers, who want to maximise the net productivity of their farms. Arable farmers want to maximise their NPP, while pastoral farmers want to maximise their NSP. The chart on the previous page shows that subsistence farming (practiced in poor and developing nations) is pretty inefficient, while intensive farming (practiced in developed nations) is ten times more productive. How is this high productivity achieved?

Intensive farming is designed to maximise productivity (crop/meat/milk etc.) by making use of any appropriate technology. The huge increase in human population over the last few hundred years has been possible due to the development of intensive farming, and most farms in the UK are intensively farmed. Intensive farming techniques to increase productivity include:

- **Selective breeding.** Most of the increases in primary productivity are due to selective breeding of crops and farm animals that grow faster and bigger (see unit 2).
- **Fertilisers*.** Primary productivity is often limited by the availability of minerals in the soil, so fertilisers overcome this limitation and increase productivity.
- **Pest Control*.** Loss of crops to pests decreases net productivity, so pest control measures increase productivity.
- **Factory Farming*.** By rearing livestock indoors and feeding them specialised diets energy losses due to heat, movement and egestion are reduced. This increases net secondary productivity.
- **Large Fields** mean less farmland is wasted with hedgerows and field margins, so overall productivity for the land is increased.
- **Monoculture** means farmers can specialise in one type of crop and find the optimum conditions for maximum productivity.
- **Mechanisation** means crops can be sown and harvested more quickly and reliably, cows can be milked more quickly and money can be saved by employing fewer farm workers.

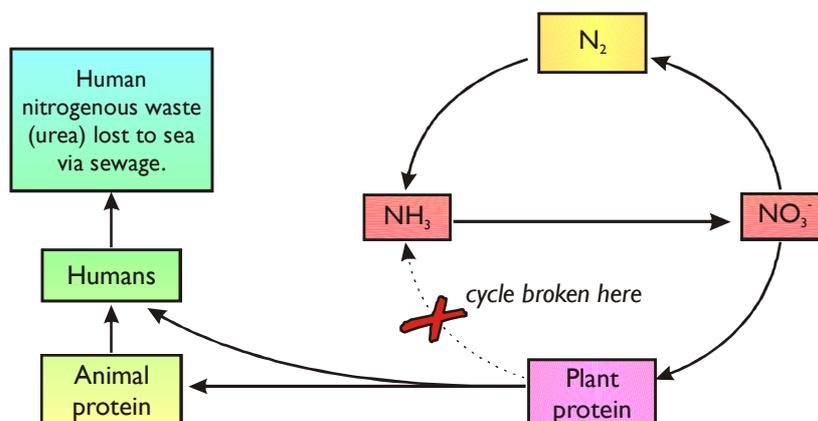
Note that some of these strategies increase net productivity by increasing gross productivity (e.g. fertilisers), while others do it by decreasing respiratory loss (e.g. by factory farming). Although these methods increase productivity there is a cost. Many of these methods require energy input from the farmer. Building livestock sheds; heating buildings, running farm machinery and producing fertiliser all require energy, usually in the form of burning fossil fuels. The farmer has to make sure that the gains in productivity outweigh the extra costs. We shall look at fertilisers, pest control and factory farming in more detail.

First, it may help to define some of the terms associated with farming:

Intensive farming	Farming designed to maximise yield of produce (crop/meat/milk etc.) by making use of any appropriate technology such as agrochemicals, machinery, etc. Most farms in the UK are intensively farmed.
Extensive farming	Farming with minimum input from the farmer. e.g. upland sheep farming.
Organic farming	Farming that uses few agrochemicals and so does not pollute the environment (3% of UK farms are organic).
Subsistence farming	Farming for the farmer's family's own needs rather than for profit. Today mainly carried out in third-world countries.
Factory farming	Raising livestock indoors in large numbers to increase productivity.
Monoculture	Growing a single crop in a field.
Polyculture	Growing many different crops in the same space, imitating the diversity of a natural ecosystem.
Arable farm	A farm that grows crops.
Pastoral farm	A farm that grows animals. The pasture is the grass that is grazed.
Mixed farm	A farm that grows plants and animals.
Agrochemicals	Collective name for chemicals applied to crops and animals, including fertilisers, pesticides, herbicides, dips, etc.
Pest	Any organism that harms crops. Can include animals, other plants or microbes.
Weed	Any plant growing in a farm that the farmer doesn't want.
Annual	A plant that lives for one year. Cereal crops are annual plants.
Perennial	A plant that lasts for many years.
Biennial	A plant that takes two years to grow from seed and die.
Nurse crop	An annual crop planted to help the main perennial crop, e.g. by shading it, providing an alternative target for pests, or improving the soil.

Fertilisers

As we've seen, minerals (like nitrate, phosphate, sulphate and potassium ions) are constantly cycled between the soil and the living organisms that live on the soil. Farming breaks the mineral cycles, since minerals taken from the soil by crops or animals are not returned to the same field, but instead transported a long distance away to feed humans. We can illustrate this for nitrogen:

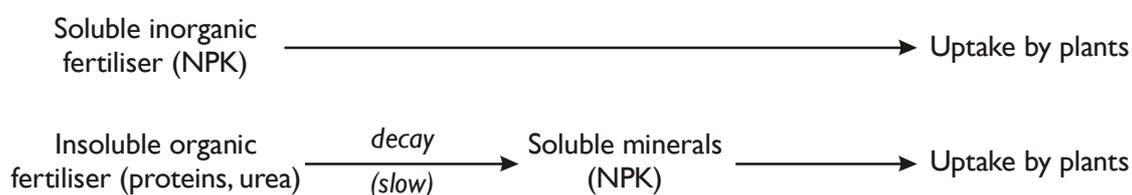


This break means that the soil is gradually depleted of minerals, so the crops don't grow as well. It applies to all minerals and is an inevitable problem of all farming (not just intensive farming). The rate of plant growth is generally limited by the availability of mineral ions in the soil, particularly nitrogen, phosphorus and potassium (NPK), so farm land has always needed to be fertilised in with these minerals some way. There are three ways this fertilisation can be done.

- **Nitrogen Fixing Crop.** A traditional way to replace lost minerals is to use a crop rotation that includes a legume crop such as clover for one year in four. During that crop's growing season, the nitrogen-fixing bacteria in the clover's root nodules make ammonia and organic nitrogen compounds from atmospheric nitrogen. Crucially, the clover is not harvested, but instead the whole crop (or sometimes just the roots) is simply ploughed back into the soil. The nitrogen that was fixed by the symbiotic bacteria in the clover's root nodules (together with the other minerals that were taken up) is thus made available to crops for the following three years. A mixture of clover and grass, called ley, does the same job for grazing pasture.
- **Inorganic Fertilisers.** Since the invention of the Haber process in 1905 it has been possible to use soluble artificial (or inorganic) fertilisers to improve yields, and this is a keystone of intensive farming. The most commonly used fertilisers are the soluble inorganic fertilisers containing nitrate, phosphate and potassium ions (NPK). Inorganic fertilisers are very effective, easy to apply, and can be tailored to each crop's individual mineral requirements, but they can also have undesirable effects on the environment. Since nitrate and ammonium ions are very soluble, they do not remain in the soil for long and are quickly leached out, ending up in local rivers and lakes and causing eutrophication. They are also

expensive and their manufacture is very energy-intensive, requiring fossil fuels, so it contributes to the greenhouse effect.

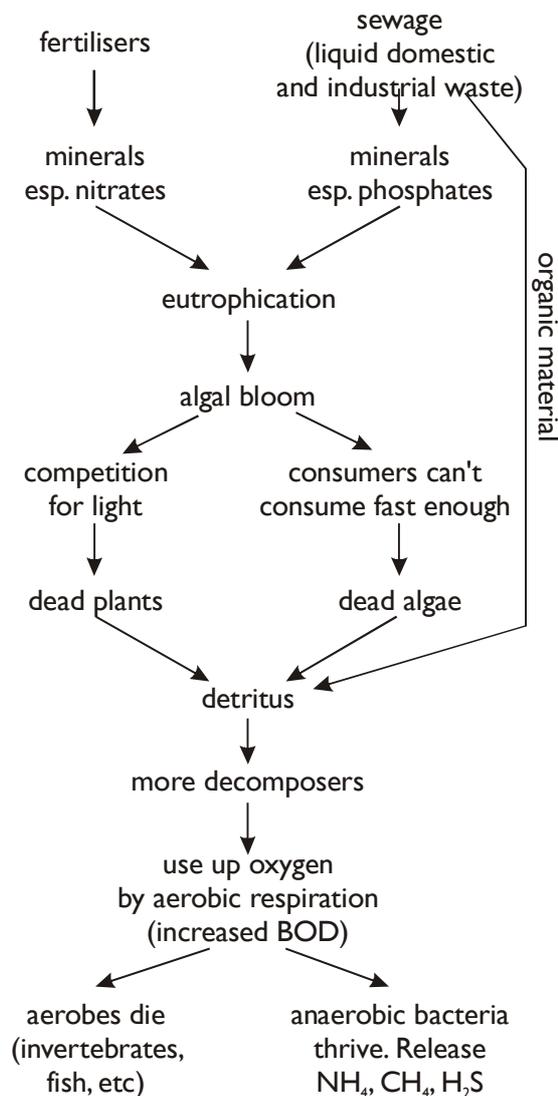
- **Organic Fertilisers.** An alternative solution, which may do less harm to the environment, is the use of natural (or organic) fertilisers, such as animal manure (farmyard manure or FYM), composted vegetable matter, crop residues, and sewage sludge. Not surprisingly, organic fertilisers are commonly just referred to as muck. They contain the main elements found in inorganic fertilisers (NPK), but contained in organic compounds such as urea, proteins, lipids and organic acids. Of course plants cannot make use of these organic materials in the soil: their roots can only take up inorganic mineral ions such as nitrate, phosphate and potassium. But the organic compounds can be digested by the soil decomposers, who then release inorganic ions that the plants can use (refer to the nitrogen cycle).



Since the compounds in organic fertilisers are less soluble than those in inorganic fertilisers, the inorganic minerals are released more slowly as they are decomposed. This prevents leaching and means they last longer. Organic fertilisers are cheap, since the organic wastes need to be disposed of anyway. Furthermore, spreading on to fields means the muck will not be dumped in landfill sites, where it may cause uncontrolled leaching. The organic material improves soil structure by binding soil particles together and provides food for soil organisms such as earthworms. This improves drainage and aeration. Some disadvantages of organic fertilisers are that they are bulky and less concentrated in minerals than inorganic fertilisers, so more needs to be spread on a field to have a similar effect, and they need heavy machinery to spread, which can damage the soil. Organic fertilisers may contain unwanted substances such as weed seeds, fungal spores and heavy metals. They are also very smelly!

Eutrophication

Eutrophication refers to the effects of nutrients on aquatic ecosystems. In particular it means a sudden and dramatic increase in nutrients due to human activity, which disturbs and eventually destroys the food web. The main causes are fertilisers leaching off farm fields into the surrounding water course, and sewage (liquid waste from houses and factories). These both contain dissolved minerals, such as nitrates and phosphates, which enrich the water.



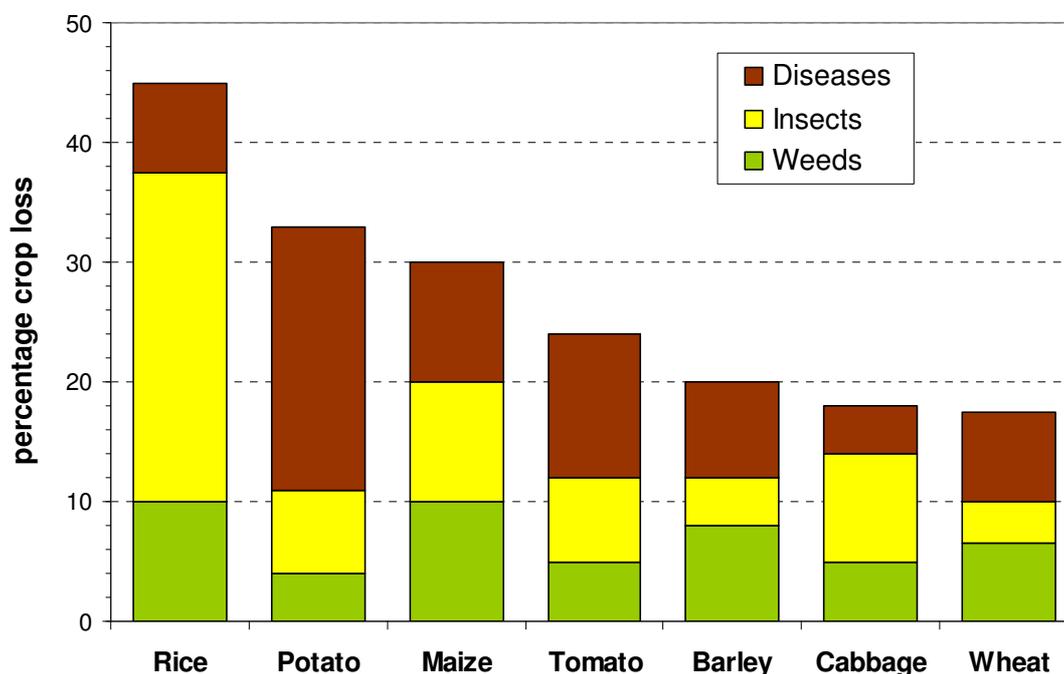
Since producer growth is generally limited by availability of minerals, a sudden increase in these causes a sudden increase in producer growth. Algae grow faster than larger plants, so they show a more obvious “bloom”, giving rise to spectacular phenomena such as red tides. Algae produce oxygen, so at this point the ecosystem is well oxygenated and fish will thrive.

However, the fast-growing algae will out-compete larger plants for light, causing the plants to die. The algae also grow faster than their consumers, so many will die without being consumed, which is not normal. These both lead to a sudden increase in detritus. Sewage may also contain organic matter, which adds to the detritus.

Microbial saprobionts can multiply quickly in response to this increase in detritus, and being aerobic they use up oxygen faster than it can be replaced by photosynthesis or diffusion from the air. The decreased oxygen concentration kills larger aerobic animals and encourages the growth of anaerobic bacteria, who release toxic waste products.

Pest Control

To farmers, a pest is any organism (animal, plant or microbe) that damages their crops. Pests can be responsible for a huge loss in crops worldwide, as this chart shows.



So **all** farmers, growers and gardeners need to use some form of pest control, or we wouldn't be able to feed the world. Pest control can be cultural (e.g. weeding or a scarecrow), chemical (e.g. pesticides) or biological (e.g. predators), and modern practice is to combine all three in integrated pest management.

Cultural Control of Pests

This refers to any farming practices that reduce the problem of pests, other than chemical or biological methods. Cultural practices include:

- Weeding – physically removing weeds and diseased crop plants to prevent reinfection.
- Crop rotation – changing the crops each year to break the life cycle of host-specific pests.
- Intercropping – planting two crops in the same field e.g. sowing rye grass with wheat encourages ladybirds to control aphids on the wheat.
- Tilling – traditional ploughing and turning of the top soil layer to bury weed seeds and expose insects to predatory birds.
- Insect barriers – e.g. sticky bands on apple tree trunks stop codling moth caterpillars.
- Beetle banks – building strips of uncultivated rough ground around and through fields. These strips are breeding grounds for beetles and other invertebrates that may predate the pest and so keep their populations under control.

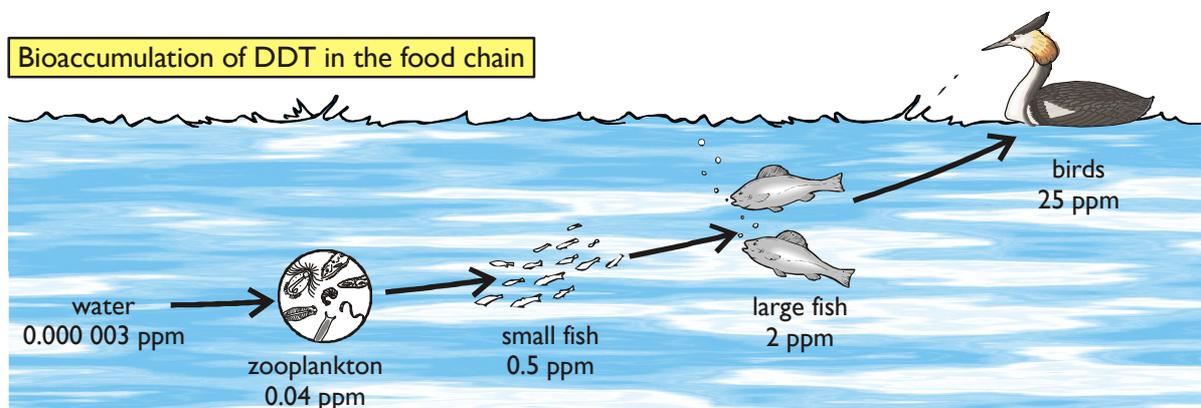
Chemical Control of Pests

Chemicals that kill pests are called pesticides. Chemical pesticides include herbicides (anti-plant chemicals); insecticides (anti-insect chemicals); fungicides (anti-fungal chemicals); and bactericides (anti-bacterial chemicals). Pesticides have been used in some form for over 1000 years, and modern intensive farming depends completely on the use of pesticides to increase yields. Some wheat crops are treated with 18 different chemicals to combat a variety of weeds, fungi and insects. In addition, by controlling pests that carry human disease, pesticides have saved millions of human lives. Good pesticides must be:

- **Selectively toxic**, which means they kill their target but not the crop or other organisms including humans. Early pesticides were non-selective (or **broad-spectrum**), which means they caused a lot of harm to the environment. Broad-spectrum pesticides can kill useful pollinating insects and pest predators, so can actually cause the pest population to increase. Modern pesticides must be selective (or **narrow-spectrum**), which is better for the environment, but they are more expensive to produce.
- **Biodegradable**, which means they are broken down by decomposers in the soil. Early pesticides were not easily broken down (they were **persistent**), so they accumulated in food chains and harmed humans and other animals, but modern pesticides biodegradable so they do not leave residues on crops.

Different kinds of pesticides are used to control different kinds of pest:

- **Insecticides**. Insects are the most important group of animal pests, like aphids and leatherjackets that eat the crop and so reduce yield. Insecticides can be **contact** or **systemic**. Contact insecticides remain on the surface of the crop and only kill insects that come into contact with it, so are not 100% effective. Systemic insecticides are absorbed into the crop and transported throughout the plant, so any insect feeding on the crop will be killed. One of the most famous insecticides is DDT, which was used very successfully from the 1940s to 80s and was responsible for eradicating malaria from southern Europe. However DDT was non-selective and persistent, so it accumulated in the food chain and killed sea birds and other top predators. DDT was banned in developed countries in 1970, and the bird populations have since recovered.



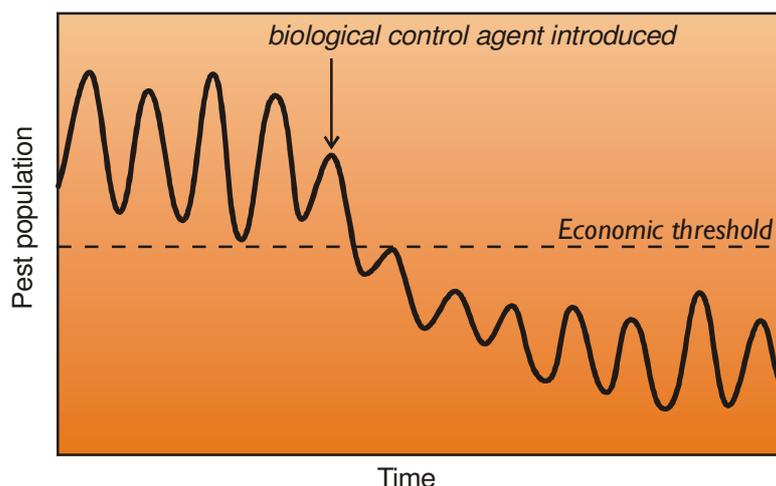
- **Herbicides**. Weeds are simply plants that the farmer (or gardener) doesn't want. Plants like wild oats, cleavers, bindweeds and thistles compete with the crop plants for light, water and minerals, and so

reduce crop yields. Weeds can also harbour pests and pathogens that can infect neighbouring crops. Weeds usually arrive on farmland by wind-dispersed seeds or they can be sown accidentally with the crop. How can a chemical kill some plants (weeds) but not others (the crop)? Fortunately, cereal crops are narrow-leaved grasses (monocotyledons), while most weeds are broad-leaved (dicotyledon) plants, and these groups have different enzymes, so herbicides can be targeted at just one group. For example the herbicide “2,4-D” is a synthetic plant hormone that causes broad-leaved plants to shoot up and die, but has no effect on cereals.

- **Fungicides.** Fungi are the most important plant pathogens, causing diseases like mildew, rusts and blackspot and rotting produce in storage. Crop seeds are often treated with fungicides before sowing.

Biological Control of Pests

As an alternative to chemical pest control, pests can be controlled using other living organisms to keep the pest numbers down – biological pest control. The organisms can be predators, parasites or pathogens, and the aim is to reduce the pest population to a level where they don't do much harm – the economic threshold. A new equilibrium should be reached where the pest and predator numbers are both kept low.



Biological pest control works particularly well when the pest has been introduced to the ecosystem and has no natural predators. An example is the cottony cushion scale insect, which was accidentally introduced to California from Australia in the late nineteenth century. In California it multiplied out of control and destroyed large numbers of citrus trees, a major Californian crop. So the ladybird beetle, one of the scale insect natural predators, was also introduced from Australia, and quickly reduced the numbers of scale insects to a safe level. Today both species coexist in California, but at low population densities.

The control species has to be chosen carefully, to ensure that it

- attacks the pest only and not other native species
- will not itself become a pest due to lack of predators or parasites
- can survive in the new environment
- does not carry disease

Control species should be trialled in a quarantine area, such as a greenhouse, before being released into the wild. If proper precautions are not taken, biological control can lead to ecological disaster. For example cane toads were introduced to Australia from Hawaii in 1935 to control beetles that feed on sugar cane crops. But the cane toads were poisonous to predators and ate a variety of prey, including native marsupials, so they are spreading through Australia and are now more of a problem than the original beetles.

Other examples include wasps controlling aphids, cactus moths controlling prickly pears, myxomatosis controlling rabbits and guppies controlling mosquitoes.

Integrated Pest Management

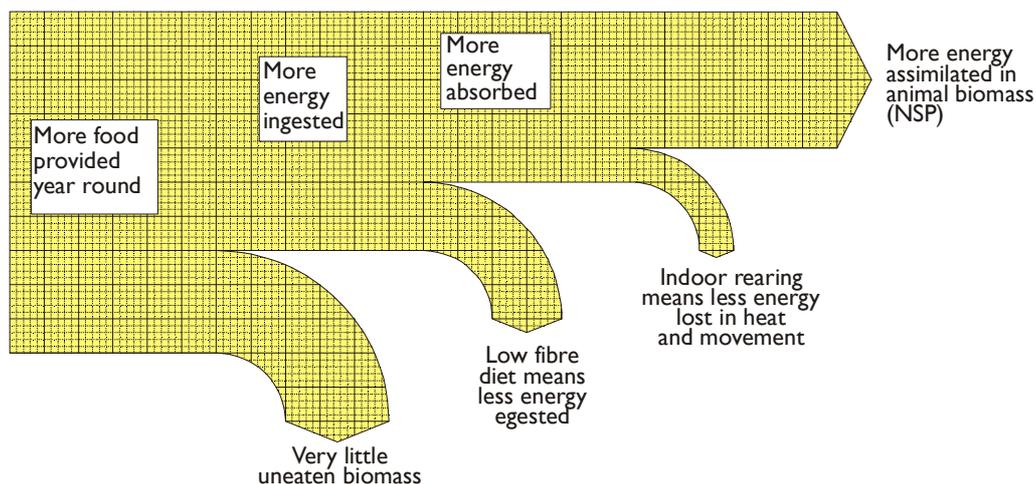
Modern intensive farming recognises the environmental dangers of the untrammelled use of pesticides, so is adopting Integrated Pest Management (IPM). IPM attempts to bring together all forms of pest management to benefit from the best of each. The aim is to reduce the effect of pesticides on the environment but without compromising the goal of maximising crop yield. There are 4 stages, each one more powerful than the one before:

1. Identify the pests and their population density at which they cause economic harm – the economic threshold. Only take action against the pest if its population is above the threshold.
2. Use suitable cultural methods to prevent pests reaching their threshold.
3. If the pest population starts to exceed threshold the use biological control to bring it down.
4. If biological control doesn't work then use chemical pesticides, but at low and carefully controlled dose, and at the best time of year to minimise damage to other organisms while maximising effect on pest.
5. At each stage the effect of that treatment is evaluated before deciding to proceed to the next stage.

Factory Farming

What about pastoral farming? How can farmers increase net secondary productivity (NSP)? The applications of intensive farming techniques to livestock are called factory farming, and include the following processes:

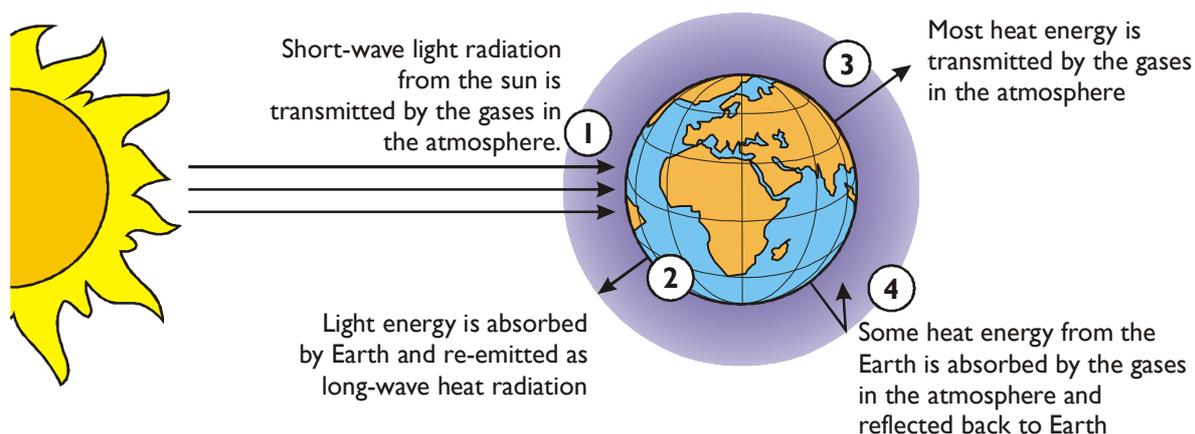
- Animals are kept indoors for part or all of the year, usually at very high density. The barn is kept warm by the collective body heat of so many animals in close proximity, and in very cold conditions buildings can be heated (though this costs the farmer). Less energy is lost as respiratory heat, so increasing NPP. In addition, animals can't move much, so they don't expend energy in muscle contraction. More of the food they eat is converted to useful biomass rather than being lost in respiration.
- Animals are given specialised, high-energy food for part or all of the year. This food has high nutritive value so animals grow quickly and can be sold sooner. The food is low in plant fibres (cellulose), so it is easy to digest and less energy is wasted in egested faeces. The food also contains mineral and vitamin supplements that the animals would normally obtain from fresh food and exposure to sunlight.
- Animals are given antibiotics to mitigate the effect of infectious disease. The dense packing of animals makes it easy for pathogens to spread from host to host, so antibiotics are essential to prevent epidemics. Antibiotics also increase growth rate by killing intestinal bacteria, though this use was banned by the EU in 2006.
- Animals are selectively bred to be fast-growing (see unit 2), and they are slaughtered before growth stops in adulthood, so the farmer doesn't waste any food, and earns profit early.
- When animals are reared outdoors their pasture is fertilised to improve the quantity and quality of grazing. This increases the animals' energy intake at little cost.



These interventions all cost money, and indeed intensive farming depends on high levels of inputs to achieve high productivity. But the gains in productivity should exceed the costs of the inputs. Factory farms produce large amounts of animal waste, which often pollute surrounding water ways. Factory farming also raises many ethical questions about the welfare of the animals. In the EU both battery cages for chickens and gestation crates for pigs are being phased out by 2012.

The Greenhouse Effect

The Earth and the moon are the same distance from the sun yet the mean temperature on Earth is 14°C and on the moon is -18°C. Why is this? It's because the Earth has an atmosphere and the moon doesn't. Certain molecules in the Earth's atmosphere keep the Earth warm by transmitting short-wave radiation from the sun, but blocking long-wave radiation from the Earth. Since this is the same way that the glass walls of a greenhouse work, this is called the greenhouse effect.

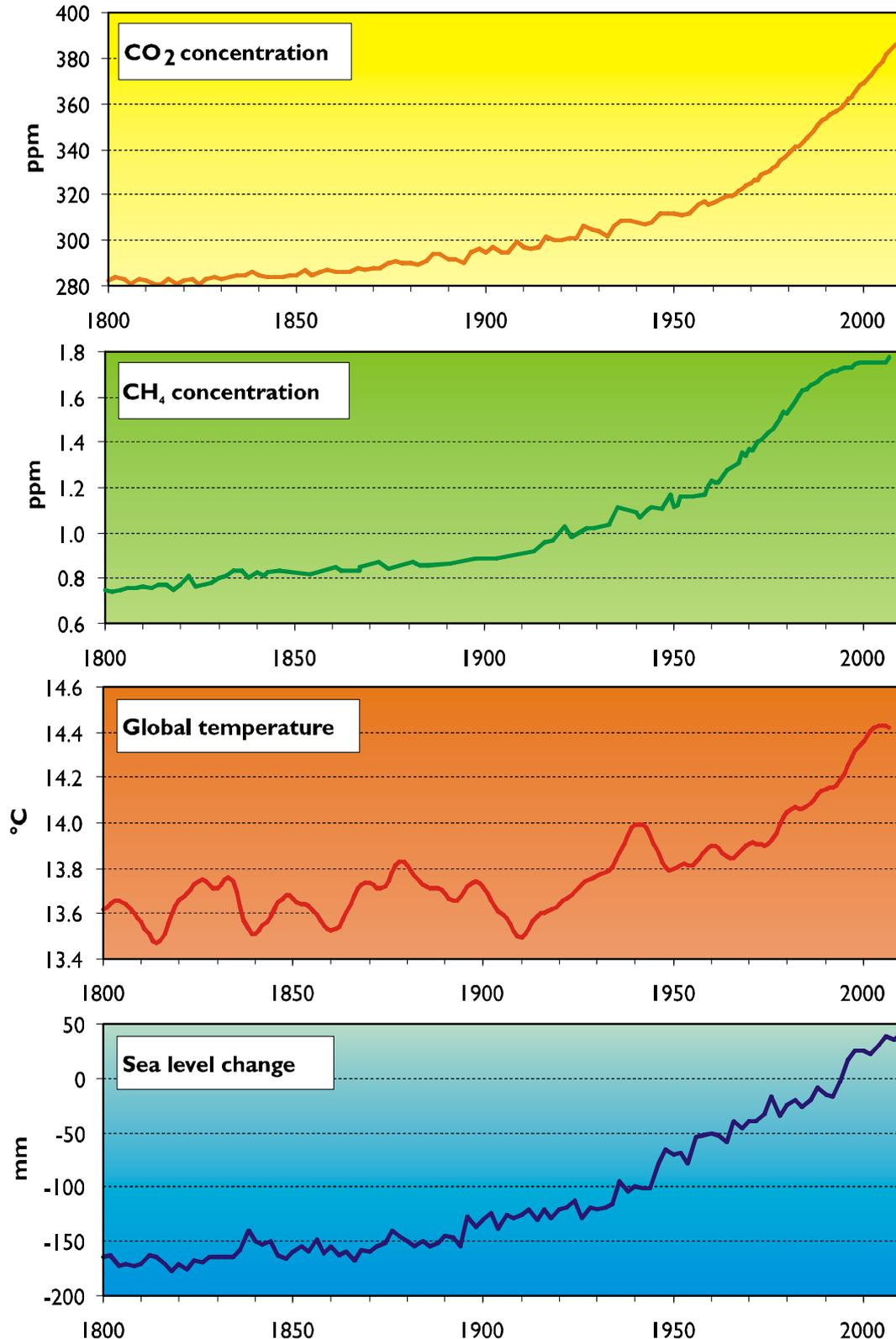


The greenhouse effect has always existed and is essential for life on Earth, as without it the temperature would be 33°C lower and there would be no liquid water. Several atmospheric gases contribute to the greenhouse effect, mainly carbon dioxide (CO₂), methane (CH₄), water vapour and ozone (O₃). The molecules of these gases all absorb radiation in the infra-red range, so are called “greenhouse gases”. The concentrations of all the gases in the atmosphere are shown in this table (ppm = parts per million):

Gas	Concentration in atmosphere (ppm)	Greenhouse gas
Nitrogen (N ₂)	780,000	
Oxygen (O ₂)	209,000	
Water vapour (H ₂ O)	~20,000	✓
Argon (Ar)	9,000	
Carbon dioxide (CO ₂)	380	✓
Neon (Ne)	18.2	
Helium (He)	5.2	
Methane (CH ₄)	1.7	✓
Krypton (Kr)	1.1	
Hydrogen (H ₂)	0.6	
Nitrous oxide (N ₂ O)	0.3	
Carbon monoxide (CO)	0.1	
Xenon (Xe)	0.09	
Ozone (O ₃)	0.04	✓

The Enhanced Greenhouse Effect

The concentrations of the greenhouse gases may be very small, but they are increasing. The result of these increases is the enhanced greenhouse effect, which is causing more heat to be trapped on Earth, leading to an increase in mean global temperature called global warming. Some of the changes in the last 200 years are shown in these charts:

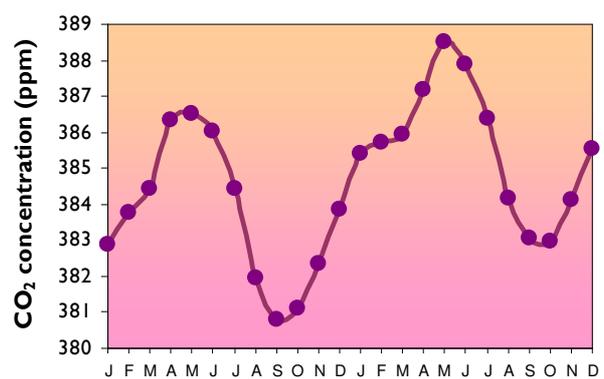
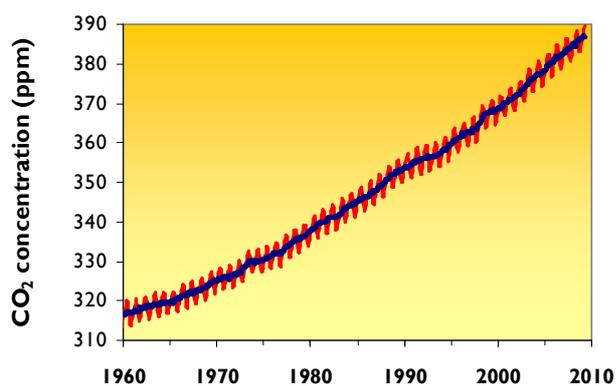


Before we look at the impact of these changes, it is worth asking where these data come from. People have been making direct measurements of temperatures and atmospheric gas concentrations at many locations over the world for over a century, but indirect (or proxy) measurements can also be made going back thousands of years. Two important techniques are:

- Tree rings. It is well-known that tree trunks have annual rings due to seasonal patterns of growth: big cells in summer followed by small cells in winter. These rings can also tell us about the climate while the tree was growing. Narrow annual rings mean that growth was slow so the temperature was colder, while wider annual rings mean that growth was faster so the temperature was warmer.
- Ice cores. The thick ice in places like Antarctica, Greenland and Siberia has been building up for tens of thousands of years, so by drilling out long ice cores many kilometres deep we can collect frozen ice from the past. These ice cores have annual rings, like trees, reflecting seasonal rainfall, so specimens from the cores can be dated. As each fresh layer of ice freezes tiny bubbles of air are trapped and using modern sensitive techniques we can analyse these ancient air bubbles for gas content. Air moves rapidly around the Earth, so these measurements represent global rather than local effects.

These studies have shown that the greenhouse gases have remained steady for ten thousand years, but then suddenly started to increase in the last 200 years.

Some of the most complete and accurate measurements of atmospheric carbon dioxide concentration are from the climate observatory at Mona Loa on Hawaii, which has been continuously monitoring atmospheric gases since the 1950's. The remote, barren, high-altitude location means measurements are undisturbed by local human activity. The carbon dioxide record shows a clear increase in the last 50 years, on top of annual fluctuations due to seasonal changes in photosynthesis and respiration. Although the Mona Loa observatory is in the southern hemisphere, these annual fluctuations reflect the northern hemisphere seasons, because that's where most of the world's plants are.



Anthropogenic Global Warming

What is causing the rise in greenhouse gases? The main sources are:

- **Burning fossil fuels.** This releases carbon dioxide that was removed from the atmosphere by plants 300 million years ago during the carboniferous era.
- **Deforestation.** This increases the carbon dioxide concentration by reducing photosynthesis and through burning or decay of the trees.
- **Agriculture.** Methane is produced by methanogenic anaerobic respiration by certain bacteria. These bacteria are found in the intestines of ruminant herbivores like cows, and in flooded rice paddy fields, where the mud is anaerobic. Since cow and rice farming have both increased dramatically in recent decades to feed the growing human population, methane emissions have also increased.
- **Landfill.** Methanogenic bacteria are also found in landfill sites, where they contribute to increased methane production.

So the changes are mainly due to the rising human population. Although greenhouse gases are also affected by natural events, like volcanic eruptions and solar activity, it seems very likely that the increases are mainly due to human activities, since the timing of the increase correlates with increased emissions due to the industrial revolution and human population increases. The changes are therefore called anthropogenic changes, since they are caused by humans. The International Panel on Climate Change (IPCC) reported in 2007 that “*the evidence for a warming trend during the 100 years ending in 2005 was unequivocal and amounted to a rise of $0.74 \pm 0.18^{\circ}\text{C}$. Human activity has very likely been the driving force for that trend*”. In other words global warming is real and at least partly anthropogenic. The IPCC also predicted that “*global temperatures will rise by a further 1.4 to 5.8°C by the end of the 21st century*”.

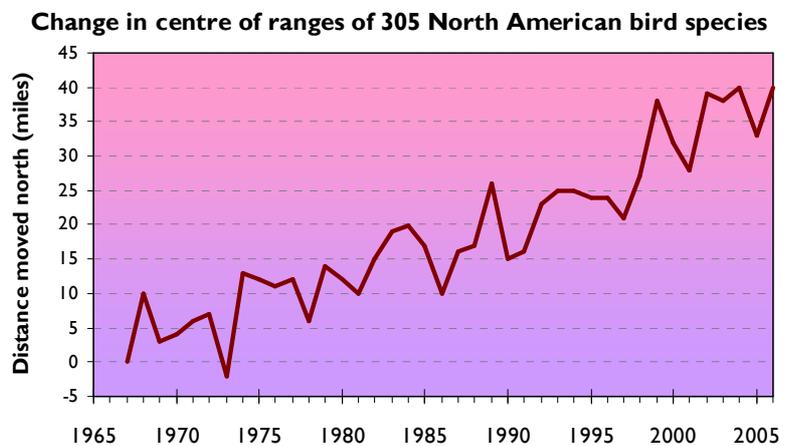
Although the mean global temperature is increasing, not everywhere will see a temperature increase: some areas may experience a cooling due to climate change or ocean current disruption. Global warming is leading to a variety of changes in the Earth, collectively called climate change. One of the biggest effects of global warming is a rise in sea level as the ice sheets on Antarctica, Greenland and Siberia begin to melt (the Arctic ice is floating, so its melting won't affect sea levels), coupled with thermal expansion of sea water. Climate change also includes changes in rainfall patterns and increased frequency of extreme weather events.

Impact of Climate Change

Predictions about the effects of global warming are difficult, but many changes have already been observed that can be attributed to anthropogenic global warming:

- **Wild animals and plants.** As habitats change, many animals and plants, who are adapted to their current environment, will struggle to survive as their environment warms up. They have to colonise new habitats with a more suitable climate or become extinct. Many species are moving towards the poles as their habitats warm and polar species like polar bears and emperor penguins are declining as their habitats disappear. Many birds are starting their annual migrations earlier and butterflies have shifted their ranges northward by 200km in Europe and North America. The white lemuroid possum, only found in the mountain forests of northern Queensland, is the first mammal species to be driven extinct by global warming.

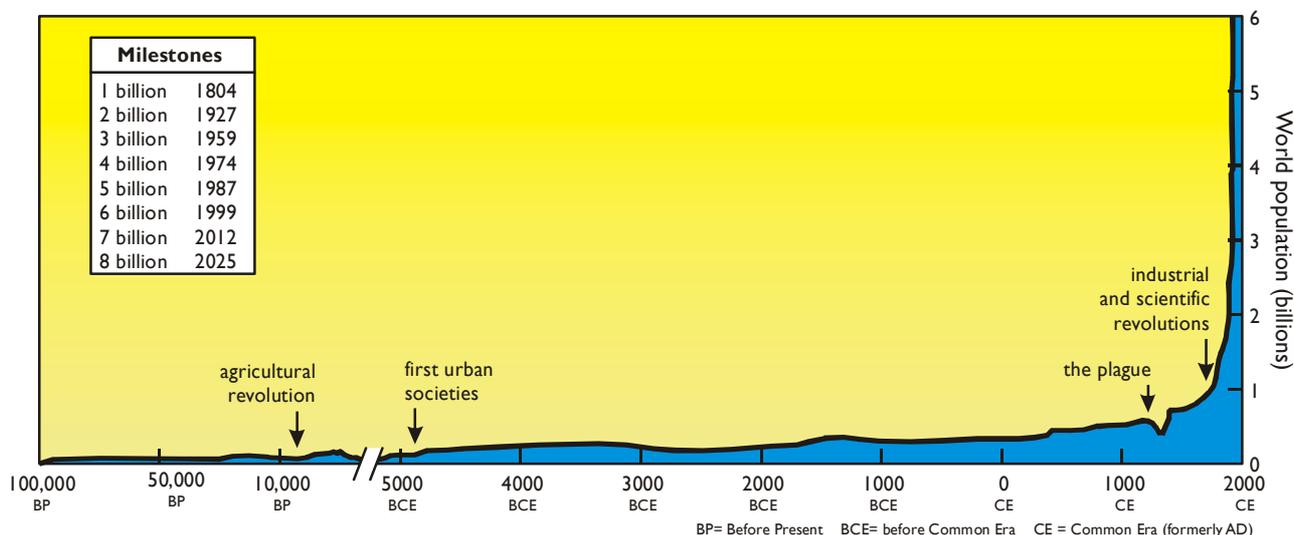
This chart shows how global warming has affected birds in North America over the last 40 years. The northward movement of the birds' ranges is consistent with the increases in annual temperature over the same time period.



- **Crops.** Increases in temperature and carbon dioxide will both contribute to an increase in growth rate in some crops, but the effects will vary around the globe, partly because the effects of global warming on rainfall are almost impossible to predict. Cool and temperate regions may be able to grow crops currently confined to warmer regions, for example barley can now be grown for the first time in Iceland. But Mediterranean and tropical regions may be unable to support their current crops as the climate becomes too warm and dry. In particular, African countries may struggle to feed their populations as staple crops fail due to rising temperatures and decreased rainfall. Globally, it is predicted that agricultural output is most likely to fall due to global warming.
- **Pests.** The geographic range of insect pests is already shifting towards the poles, as mild winters mean that insects can now survive in areas where they couldn't before. For example, the green shield bug, a common pest of vegetable crops in Mediterranean countries, has recently been found in southern England for the first time. Since insects like mosquitoes are also vectors of human and animal pathogens, tropical diseases could soon be found in more northern countries. In 2007, bluetongue, a viral disease of cattle and sheep carried by a midge vector, appeared for the first time in Britain.

Human Populations

Homo sapiens evolved around 400,000 years ago, and for most of that time the total world population was below 100,000 (about the size of a small town like Darlington today). The population started to increase following the transition to farming (the agricultural revolution) 10,000 years ago, but really accelerated following the industrial and scientific revolutions starting in Europe in the 18th century.



The population of humans today is increasing by 2.5 people every second (or by the population of New York every month), and is predicted to reach 7 billion in 2012. The increase in population, or growth rate, depends on four factors:

$$\text{growth rate} = (\text{birth rate} - \text{death rate}) + (\text{immigration rate} - \text{emigration rate})$$

The equation shows that growth rate can increase by increasing the birth rate or decreasing the death rate (ignoring migration). The staggering human population growth over the last two centuries is entirely due to a massively decreased death rate caused by the improvements in farming described earlier, and in medicine. The increased growth rate has therefore happened at different times for different countries.

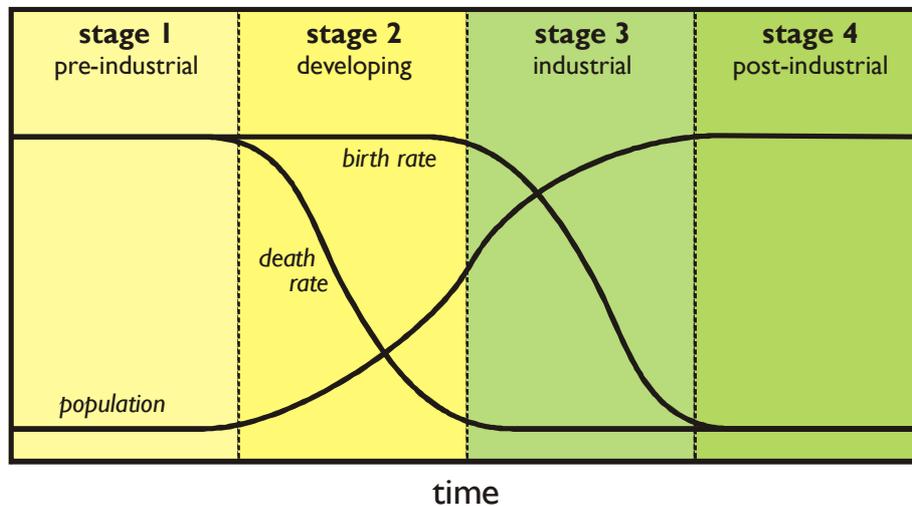
Growth rate can also be expressed as a percentage change:

$$\text{annual percent change} = \frac{(\text{population at end of year} - \text{population at start of year})}{\text{population at start of year}} \times 100$$

If the population decreases the population change will be negative.

The Demographic Transition

History has shown that the birth and death rates of a country change in particular ways as the country develops. These changes are called the demographic transition, and there are four stages:



stage 1
pre-industrial

In pre-industrial societies, birth and death rates are both high. The lack of medicine and poor sanitation means that child mortality is high and life expectancy is short. Children work from a young age, so are useful and cheap to bring up, so parents choose to have many children. Countries in sub-Saharan Africa, such as Angola and Ethiopia are still stage 1 societies.

stage 2
developing

In developing societies the death rate (especially child mortality) decreases due to improved farming, nutrition, health care, sanitation and education. Life expectancy increases, but birth rate remains high, so the population increases. Most European countries went through stage 2 in the 18th century, but developing countries such as Afghanistan and Laos are in stage 2 today.

stage 3
industrial

In industrial societies the death rate is low and the birth rate starts to fall as parents choose to have fewer children. This family planning often results from urbanisation, so children are no longer needed to work the land, and the cost of their upbringing and education increases. The population still increases, but at a slower rate. Many countries in the world today are in stage 3 including Mexico, India, Pakistan, Saudi Arabia and South Africa.

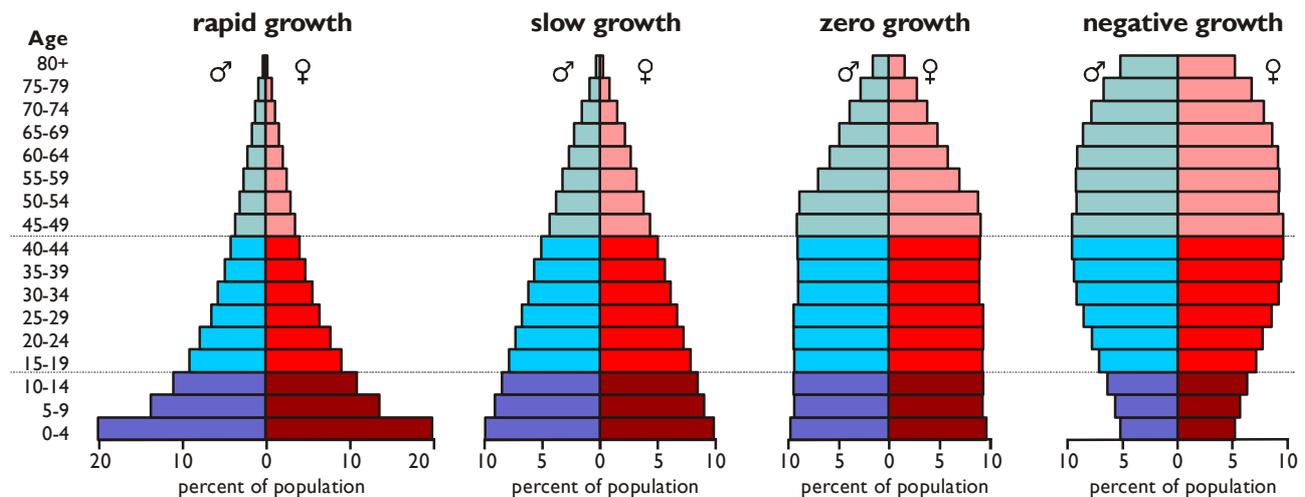
stage 4
post-industrial

In post-industrial societies the birth and death rate are both low, so the population is stable, but high. Countries like the United States, Canada, Australia and most of Europe are in stage 4. In some countries (such as the UK, Germany and Japan) the birth rate falls below the death rate, so the population starts to decrease. This decrease is sometimes described as stage 5.

Age Structure

The demographic changes also lead to changes in life expectancy and the age structure of a population, i.e. the proportions of young and old people. These changes can be shown as population pyramids and survivorship curves.

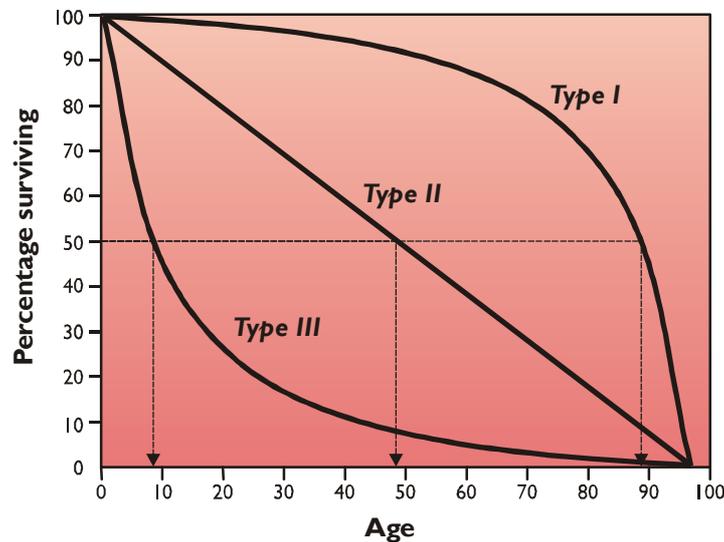
Population pyramids (or age pyramids) show the population of a country as a bar chart with a bar for each age group (usually 5 or 10 years per bar). Males (σ) and females (ρ) are usually shown separately on the two sides of the chart. To interpret these charts, it helps to put the ages into three bands: pre-reproductive (0–14); reproductive (15–44) and post-reproductive ages (over 45).



The shape of an age pyramid tells us about the future growth of the population and about life expectancy.

- The wider the base of the pyramid, the faster the population growth. Imagine the wide bottom bars moving up the pyramid over time into the reproductive age band. A pyramid with a narrow base indicates a falling population.
- The steeper the pyramid, the longer the life expectancy. An age pyramid with a wide base that declines quickly and has a narrow tip indicates high infant mortality and a short life expectancy. A more rectangular shape with a broader tip indicates long life expectancy.

Survivorship curves (or survivor curves) are constructed by tracking a group of individuals born around the same time (a cohort) from birth until the last one has died, and recording each individual's age at death. The percentage of the cohort surviving is plotted at each age. So all survivorship curves start at 100% and finish at 0%, but have different shapes in between. Survivorship curves allow us to calculate life expectancy – the mean lifespan of a cohort – by simply reading off the age at which 50% survive.



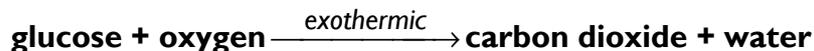
Three kinds of shape are found, illustrated by the three idealised curves on the graph.

- In **type I curves** there is a long life expectancy, with low infant mortality and most of the cohort dying in old age. Type I curves are shown by large mammals and Industrial human societies, where families are small and there is a high investment in parental care.
- In **type II curves** there is an intermediate life expectancy and a roughly constant death rate regardless of age. Type II curves are shown by animals that are equally susceptible to predation or disease at any age, such as small mammals and many birds. They are also shown by human societies facing a serious epidemic, such as the AIDS epidemic in Botswana.
- In **type III curves**, there is a short life expectancy, with most of the cohort dying in infancy and few surviving to old age. Type III curves are shown by animals that do little or no parenting and produce large numbers of offspring to compensate, such as insects and fish. They are also shown by pre-industrial human societies with poor healthcare and high infant mortality.

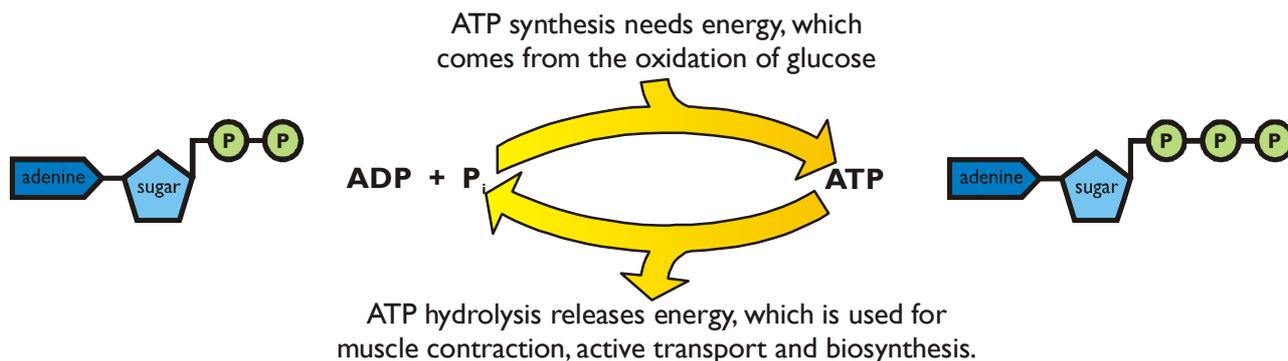
These three curves represent extreme cases, and in practice many countries show intermediate curves.

Respiration

All living cells require energy, and this energy is provided by the oxidation of glucose – respiration.



The oxidation of glucose is strongly exothermic, but in respiration the energy is released not as heat, but in the form of chemical energy in a compound called **ATP** (adenosine triphosphate). ATP is built up from ADP and phosphate (PO_4^{3-} , abbreviated to P_i). So all respiration really does is convert chemical energy stored in glucose into chemical energy stored in ATP.



ATP is a nucleotide (one of the four found in DNA), but it also has this other function as an energy storage molecule. So ATP is actually a bigger molecule than glucose, but it is very soluble and the energy it contains can be released very quickly and easily. As we'll see later, over 30 molecules of ATP can be made from each glucose molecule in respiration, so ATP stores a much smaller amount of energy than glucose. This is a good thing, as these small packets of easily-released energy are more useful to cells and can be used to do simple common jobs, as the next paragraph shows. An analogy would be that small change (ATP) is often more useful than large bank notes (glucose).

What is the energy in ATP used for?

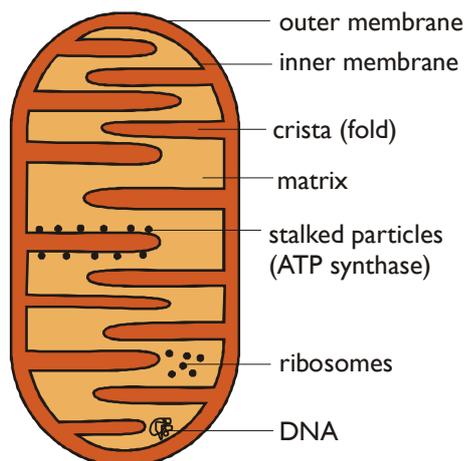
The processes in a cell that require energy can be put into three groups:

- **Muscle contraction** and other forms of movement, such as cilia, flagella, cytoplasmic streaming, etc. Each step of the muscle crossbridge cycle costs one ATP molecule.
- **Active transport.** Each shape change in an active transport protein pump costs one ATP molecule.
- **Biosynthesis** – building up large molecules from smaller ones, e.g. protein synthesis, DNA replication, starch synthesis, etc. Each monomer added to a growing polymer chain costs one ATP molecule.

Since these processes use ATP, they all involve ATPase enzymes. ATPases catalyse the hydrolysis of ATP to $\text{ADP} + \text{P}_i$, and do work with the energy released.

All the thousands of chemical reactions taking place in a cell are referred to as Metabolism. To make the reactions easier to understand, biochemists arrange them into metabolic pathways. The intermediates in these metabolic pathways are called metabolites.

Mitochondria



Much of respiration takes place in the mitochondria. Mitochondria have a double membrane: the outer membrane contains many protein channels called porins, which let almost any small molecule through; while the inner membrane is more normal and is selectively permeable to solutes. The inner membrane is highly folded into projections called cristae, giving a larger surface area. The electron microscope reveals blobs on the inner membrane, called stalked particles. These blobs have now been identified as enzyme complexes that synthesise ATP, and are more correctly called ATP synthase enzymes (more later). The space inside the

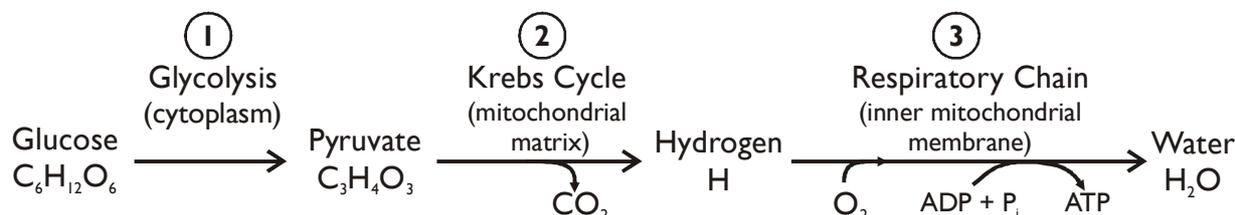
inner membrane is called the matrix, and is where the Krebs cycle takes place. The matrix also contains DNA, tRNA and ribosomes, and some genes are replicated and expressed here.

Details of Respiration

The equation for cellular respiration is usually simplified to:



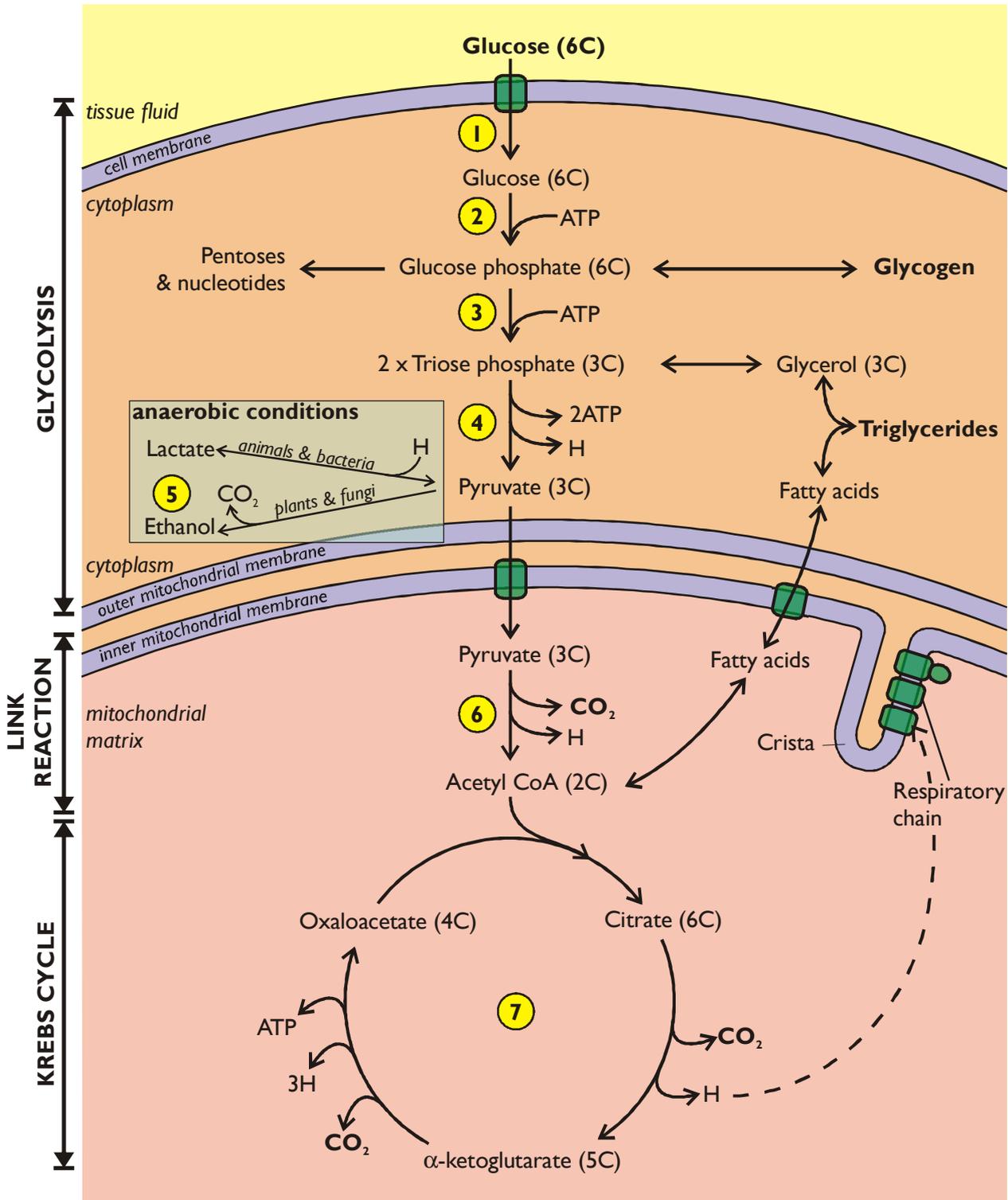
But in fact this is a summary of a complex metabolic pathway, comprising at least 30 separate steps. To understand respiration in detail we can first break it up into 3 stages:



Before we look at these stages in detail, there are a few points to note from this summary.

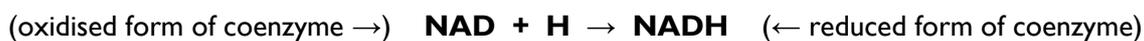
- The different stages of respiration take place in different parts of the cell. This compartmentation allows the cell to keep the various metabolites separate, and to control the stages more easily.
- The energy released by respiration is in the form of ATP.
- Since this summarises so many separate steps (often involving H^+ and OH^- ions from the solvent water), it is meaningless to try to balance the summary equation.
- The release of carbon dioxide takes place before oxygen is involved. It is therefore not true to say that respiration turns oxygen into carbon dioxide; it is more correct to say that respiration turns glucose into carbon dioxide, and oxygen into water.
- Stage 1 (glycolysis) is anaerobic respiration, while stages 2 and 3 are the aerobic stages.

Glycolysis and the Krebs Cycle



1. Glucose enters cells from the tissue fluid by facilitated diffusion using a specific glucose carrier. This carrier can be controlled (gated) by hormones such as insulin, so that uptake of glucose can be regulated.

- The first step is the phosphorylation of glucose to form glucose phosphate, using phosphate from ATP. There are two reasons for this step. Firstly, it keeps glucose in the cell by effectively removing “pure” glucose, so glucose will always diffuse down its concentration gradient from the tissue fluid into the cell (glucose phosphate no longer fits the membrane carrier). Secondly, it “activates” glucose for biosynthesis reactions: glucose phosphate is the starting material for the synthesis of pentose sugars (and therefore nucleotides and DNA), glycogen and starch.
- Glucose is phosphorylated again (using another ATP) and split into two triose phosphate (3 carbon) sugars. From now on everything happens twice per original glucose molecule.
- The triose sugar is changed over several steps to form pyruvate, a 3-carbon compound. In these steps some energy is released to form ATP (the only ATP formed in glycolysis), and a hydrogen atom is also released. This hydrogen atom is very important as it stores energy, which is later used by the respiratory chain to make more ATP. The hydrogen atom is taken up and carried to the respiratory chain by the coenzyme NAD, which becomes reduced in the process.



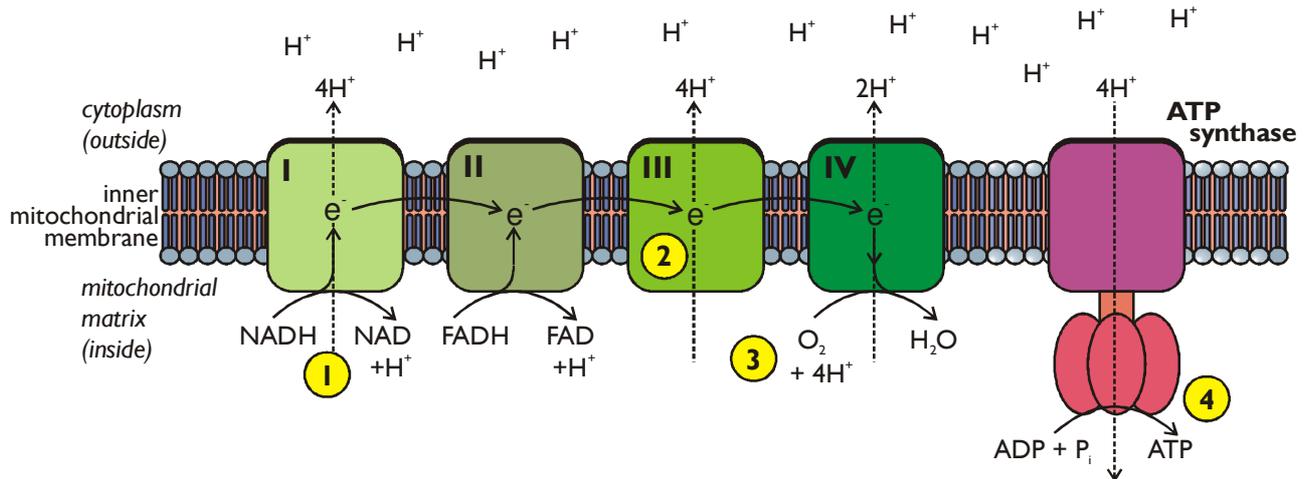
Pyruvate marks the end of glycolysis, the first stage of respiration. Pyruvate can also be turned back into glucose by reversing glycolysis, and this is called gluconeogenesis.

- In the absence of oxygen pyruvate is converted into lactate or ethanol in anaerobic respiration (p 62).
- In the presence of oxygen pyruvate enters the mitochondrial matrix to proceed with aerobic respiration. Once pyruvate has entered the inside of the mitochondria (the matrix), it is converted to a compound called acetyl coA. Since this step links glycolysis and the Krebs Cycle, it is referred to as the link reaction. In this reaction pyruvate loses a CO₂ and a hydrogen to form a 2-carbon acetyl compound, which is temporarily attached to another coenzyme called coenzyme A (or just coA), so the product is called acetyl coA. The CO₂ diffuses through the mitochondrial and cell membranes by lipid diffusion, out into the tissue fluid and into the blood, where it is carried to the lungs for removal. The hydrogen is taken up by NAD again.
- The acetyl CoA then enters the Krebs Cycle, named after Sir Hans Krebs, who discovered it in the 1940s at Sheffield University. It is one of several cyclic metabolic pathways, and is also known as the citric acid cycle or the tricarboxylic acid cycle. The 2-carbon acetyl is transferred from acetyl coA to the 4-carbon oxaloacetate to form the 6-carbon citrate. Citrate is then gradually broken down in several steps to re-form oxaloacetate, producing carbon dioxide and hydrogen in the process. Some ATP is also made directly in the Krebs cycle. As before, the CO₂ diffuses out the cell and the hydrogen is taken up by NAD, or by an alternative hydrogen carrier called FAD. These hydrogen atoms are carried to the inner mitochondrial membrane for the final part of respiration.

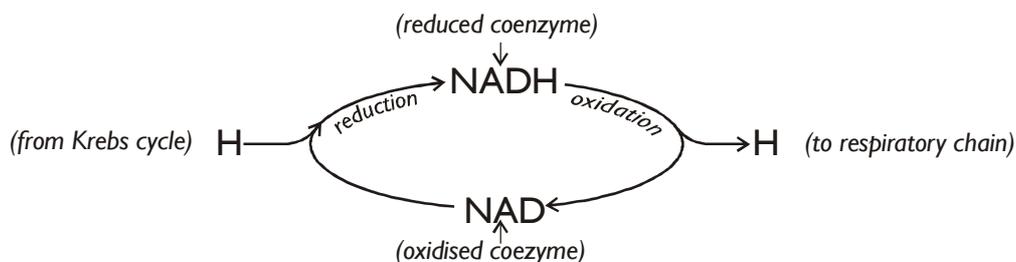
The Respiratory Chain

The respiratory chain (or electron transport chain, or oxidative phosphorylation) is an unusual metabolic pathway in that it takes place within the inner mitochondrial membrane, using integral membrane proteins. There are five of these proteins, called I, II, III, IV and ATP synthase. They each contain up to 40 individual polypeptide chains, which perform many different functions including enzymes and trans-membrane pumps.

In the respiratory chain the hydrogen atoms from NADH gradually release all their energy to form ATP, and are finally combined with oxygen to form water.



1 NADH molecules bind to protein I and release their hydrogen atoms as protons (H^+) and electrons (e^-). FADH binds to complex II rather than complex I to release its hydrogen. The NAD and FAD molecules then return to the Krebs Cycle to collect more hydrogen, so these coenzymes are constantly shuttling between their oxidised and reduced forms:



- The electrons are passed along the chain of proteins. The energy of the electrons is used to pump protons across the inner mitochondrial membrane by active transport through the proteins, forming a proton gradient across the membrane.
- Finally, the electrons are combined with protons and molecular oxygen (O_2) to form water, the final end-product of respiration. The oxygen diffused in from the tissue fluid, crossing the cell and mitochondrial membranes by lipid diffusion. Oxygen is only involved at the very last stage of respiration as the final electron acceptor, but without it the whole respiratory chain stops.
- The energy of the electrons is now stored in the form of the proton gradient across the inner mitochondrial membrane. It's a bit like using energy to pump water uphill into a high reservoir, where it

is stored as potential energy. And just as the potential energy in the water can be used to generate electricity in a hydroelectric power station, so the potential energy in the proton gradient can be used to generate ATP in the ATP synthase enzyme. The ATP synthase enzyme has a proton channel through it, and as the protons “fall down” this channel their energy is used to make ATP, spinning the globular head as they go. It takes 4 protons to synthesise 1 ATP molecule.

This synthesis of ATP is called oxidative phosphorylation because it uses oxygen to phosphorylate ADP. The method of storing energy by creating a proton gradient across a membrane is called chemiosmosis, and was discovered by Peter Mitchell in the 1960s, for which work he got a Nobel prize in 1978. Some poisons act by making proton channels in mitochondrial membranes, so giving an alternative route for protons and stopping the synthesis of ATP. This also happens naturally in the brown fat tissue of new-born babies and hibernating mammals: respiration takes place, but no ATP is made, with the energy being turned into heat instead.

Summary of Oxidative Phosphorylation

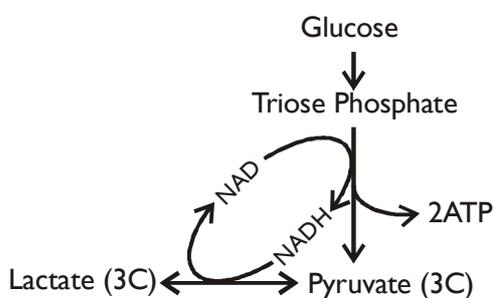
1. NADH releases its H and is oxidised to NAD, which returns to the Krebs cycle ($\text{NADH} \rightarrow \text{NAD} + \text{H}^+ + \text{e}^-$).
2. The electron is passed along the chain of proteins in the inner mitochondrial membrane, releasing its energy as it goes.
3. Oxygen combines with hydrogen to form water ($\text{O}_2 + \text{H}^+ + \text{e}^- \rightarrow \text{H}_2\text{O}$).
4. The energy of the electron is used to make ATP in the ATP synthase enzyme ($\text{ADP} + \text{P}_i \rightarrow \text{ATP}$).

Anaerobic Respiration

If there is no oxygen (anaerobic conditions) then the final reaction to make water cannot take place, no electrons can leave the respiratory chain, and so NADH cannot unload any hydrogens to the respiratory chain. This means that there is no NAD in the cell; it's all in the form of NADH. Without NAD as a coenzyme, some of the enzymes of the Krebs cycle and glycolysis cannot work, so the whole of respiration stops.

Some cells can get round this problem using anaerobic respiration. This adds an extra step to the end of glycolysis that regenerates NAD, so allowing glycolysis to continue and some ATP to be made. Anaerobic respiration only makes 2 ATPs per glucose, but it's better than nothing! There are two different kinds of anaerobic respiration:

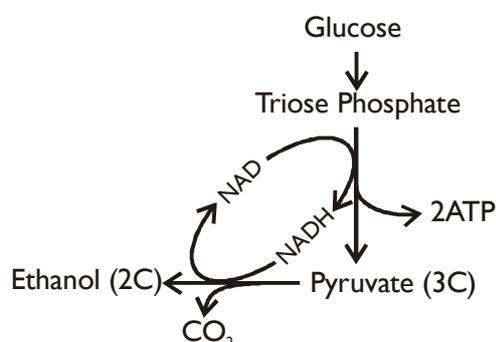
Lactic Acid Anaerobic Respiration



In animals and bacteria the extra step converts pyruvate to lactate (or lactic acid). This is a reduction, so NADH is used and NAD is regenerated, to be used in glycolysis. The reaction is reversible, so the energy remaining in the lactate molecule can be retrieved when oxygen becomes available and the lactate is oxidised via the rest of aerobic respiration.

The bacteria used to make yogurt use this reaction, as do muscle cells and red blood cells in humans. Unfortunately the lactate is poisonous, causing acidosis in muscles cells, which stops enzymes working and causes muscle fatigue and cramp. So anaerobic respiration in muscles cannot be continued for very long.

Ethanolic Anaerobic Respiration



In plants and fungi the extra steps converts pyruvate to ethanol. This is also a reduction, so NADH is used and NAD is regenerated, to be used in glycolysis. Ethanol is a two-carbon compound and carbon dioxide is also formed. This means the reaction is irreversible, so the energy in the ethanol cannot be retrieved by the cells.

Ethanolic anaerobic respiration is also known as fermentation, and we make use of fermentation in yeast to make ethanol in beer and wine.

How Much ATP is made in Respiration?

We can now summarise respiration and see how much ATP is made from each glucose molecule. ATP is made in two different ways:

- Some ATP molecules are made directly by the enzymes in glycolysis or the Krebs cycle. This is called substrate level phosphorylation (since ADP is being phosphorylated to form ATP).
- Most of the ATP molecules are made by the ATP synthase enzyme in the respiratory chain. Since this requires oxygen it is called oxidative phosphorylation. Scientists don't yet know exactly how many protons are pumped in the respiratory chain, but the current estimates are: 10 protons pumped by NADH; 6 by FADH; and 4 protons needed by ATP synthase to make one ATP molecule. This means that each NADH can make 2.5 ATPs (10/4) and each FADH can make 1.5 ATPs (6/4). Previous estimates were 3 ATPs for NADH and 2 ATPs for FADH, and these numbers still appear in most textbooks, although they are now probably wrong.

Two ATP molecules are used at the start of glycolysis to phosphorylate the glucose, and these must be subtracted from the total.

The table below is an "ATP account" for aerobic respiration, and shows that 32 molecules of ATP are made for each molecule of glucose used in aerobic respiration. This is the maximum possible yield; often less ATP is made, depending on the circumstances. Anaerobic respiration only produces the 2 molecules of ATP from the first two rows. You don't need to learn this table, but you should understand it.

Stage	molecules produced per glucose	Final ATP yield (old method)	Final ATP yield (new method)
Glycolysis	2 ATP used	-2	-2
	4 ATP produced (2 per triose phosphate)	4	4
	2 NADH produced (1 per triose phosphate)	6	5
Link Reaction	2 NADH produced (1 per pyruvate)	6	5
Krebs Cycle	2 ATP produced (1 per acetyl coA)	2	2
	6 NADH produced (3 per acetyl coA)	18	15
	2 FADH produced (1 per acetyl coA)	4	3
Total		38	32

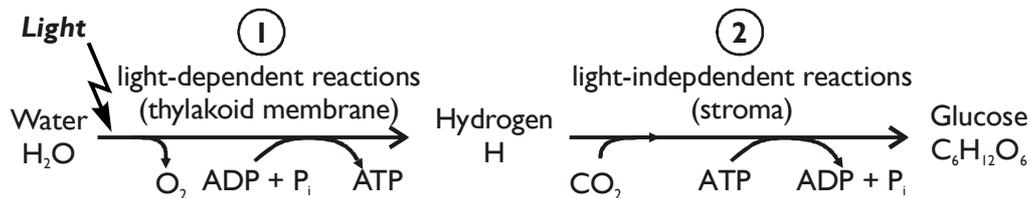
Other substances can also be used to make ATP. Glycogen of course is the main source of glucose in humans. Triglycerides are broken down to fatty acids and glycerol, both of which enter the Krebs Cycle. A typical triglyceride molecule might make 50 acetyl CoA molecules, yielding 500 ATP molecules. Fats are thus a very good energy store, yielding 2.5 times as much ATP per g dry mass as carbohydrates. Proteins are not normally used to make ATP, but in starvation they can be broken down and used in respiration. They are first broken down to amino acids, which are converted into pyruvate and Krebs Cycle metabolites and then used to make ATP.

Photosynthesis

Photosynthesis is essentially the reverse of respiration. It is usually simplified to:



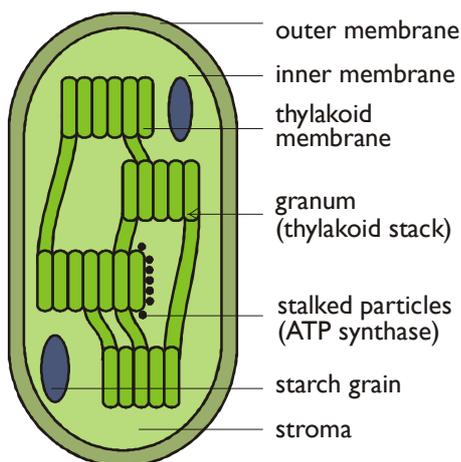
But again this simplification hides numerous separate steps. To understand photosynthesis in detail we can break it up into 2 stages:



- The light-dependent reactions use light energy to split water and make ATP, oxygen and energetic hydrogen atoms. This stage takes place within the thylakoid membranes of chloroplasts, and is very much like the respiratory chain, only in reverse.
- The light-independent reactions don't need light, but do need the products of the light-dependent stage (ATP and H), so they stop in the absence of light. This stage takes place in the stroma of the chloroplasts and involves the fixation of carbon dioxide and the synthesis of glucose.
- Like respiration, oxygen and carbon dioxide are quite separate. Plants do not turn carbon dioxide into oxygen; they turn carbon dioxide into glucose, and water into oxygen.

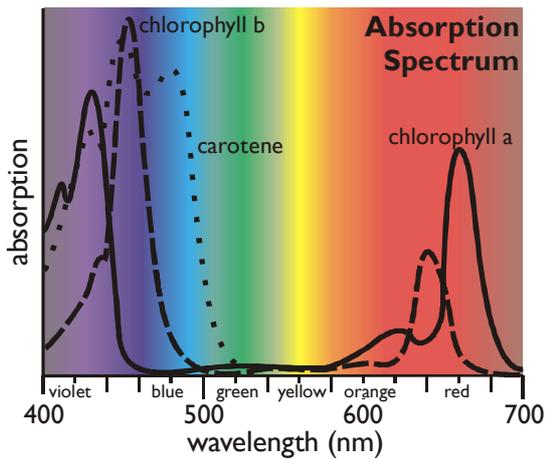
We shall see that there are many similarities between photosynthesis and respiration, and even the same enzymes are used in some steps.

Chloroplasts



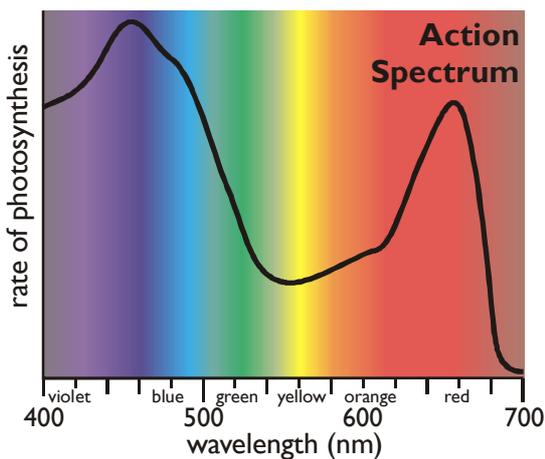
Photosynthesis takes place entirely within chloroplasts. Like mitochondria, chloroplasts have a double membrane, but in addition chloroplasts have a third membrane called the thylakoid membrane. This is folded into thin vesicles (the thylakoids), enclosing small spaces called the thylakoid lumen. The thylakoid vesicles are often layered in stacks called grana. The thylakoid membrane contains the same ATP synthase particles found in mitochondria. Chloroplasts also contain DNA, tRNA and ribosomes, and they often store the products of photosynthesis as starch grains and lipid droplets.

Chlorophyll



Chloroplasts contain two different kinds of chlorophyll, called chlorophyll a and b, together with a number of other light-absorbing accessory pigments, such as the carotenoids and luteins (or xanthophylls). These different pigments absorb light at different wavelengths, so having several different pigments allows more of the visible spectrum to be used. The absorption spectra of pure samples of some of these pigments are shown in the graph on the left. A low absorption means that those wavelengths are not absorbed and so cannot be used, but instead are reflected or

transmitted. Different species of plant have different combinations of photosynthetic pigments, giving rise to different coloured leaves. In addition, plants adapted to shady conditions tend to have a higher concentration of chlorophyll and so have dark green leaves, while those adapted to bright conditions need less chlorophyll and have pale green leaves.

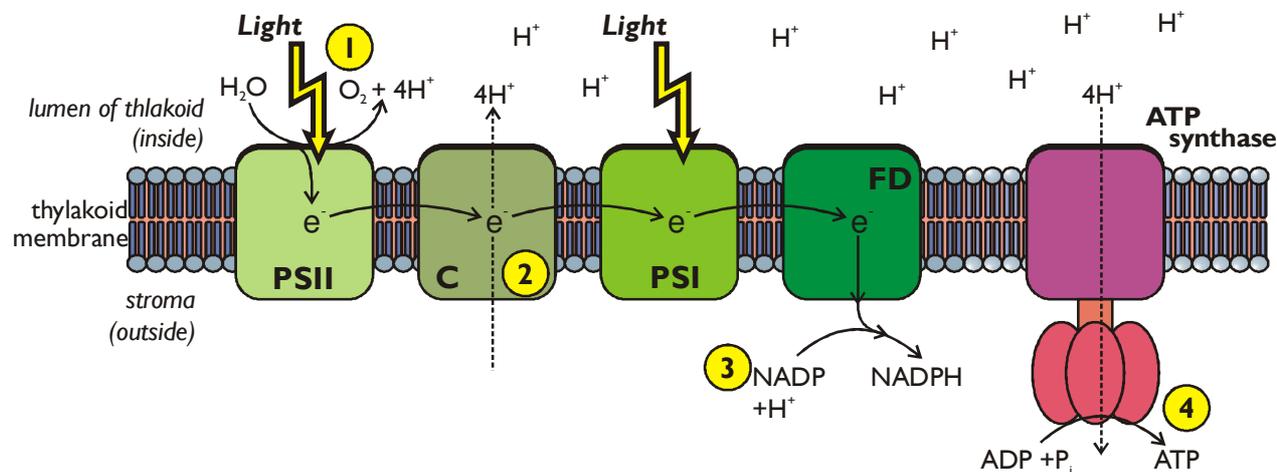


By measuring the rate of photosynthesis using different wavelengths of light, an action spectrum is obtained. The action spectrum can be well explained by the absorption spectra above, showing that these pigments are responsible for photosynthesis.

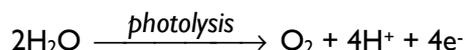
Chlorophyll is a fairly small molecule (not a protein) with a structure similar to haem, but with a magnesium atom instead of iron. Chlorophyll and the other pigments are arranged in complexes with proteins, called photosystems. Each photosystem contains some 200 chlorophyll molecules and 50 molecules of accessory pigments, together with several protein molecules (including enzymes) and lipids. These photosystems are located in the thylakoid membranes and they hold the light-absorbing pigments in the best position to maximise the absorbance of photons of light. The chloroplasts of green plants have two kinds of photosystem called photosystem I (PSI) and photosystem II (PSII). These absorb light at different wavelengths and have slightly different jobs in the light dependent reactions of photosynthesis, as we shall see.

The Light-Dependent Reactions

The light-dependent reactions (or photophosphorylation) take place on the thylakoid membranes using four membrane-bound proteins: photosystem I (PSI), photosystem II (PSII), cytochrome (C) and ferredoxin (FD). In these reactions light energy is used to split water, oxygen is given off, hydrogen is produced and ADP is phosphorylated to make ATP.

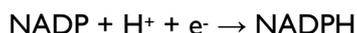


1. Chlorophyll molecules in PSII absorb photons of light, exciting chlorophyll electrons to a higher energy level and causing a charge separation within PSII. This charge separation drives the splitting (or photolysis) of water molecules to make oxygen (O_2), protons (H^+) and electrons (e^-):



Water is a very stable molecule and it requires the energy from 4 photons of light to split one water molecule. The oxygen produced diffuses out of the chloroplast and eventually into the air; the protons build up in the thylakoid lumen causing a proton gradient; and the electrons from water replace the excited electrons that have been ejected from chlorophyll.

2. The excited, high-energy electrons are passed along the chain of proteins in the thylakoid membrane, in a similar way to the respiratory chain. In PSI more light energy is absorbed and passed to the electron, which gains energy as it goes. The energy of the electrons is used to pump protons from stroma to lumen, creating a proton gradient across the thylakoid membrane.
3. Finally, the electron is recombined with a proton to form a hydrogen atom, which is taken up by the coenzyme NADP, reducing it to NADPH.



Note that while respiration uses the coenzyme NAD to carry hydrogen, photosynthesis always uses its close relative, NADP.

4. The combination of the water splitting and proton pumping causes a proton gradient across the thylakoid membrane. This gradient is used to make ATP using the ATP synthase enzyme in exactly the

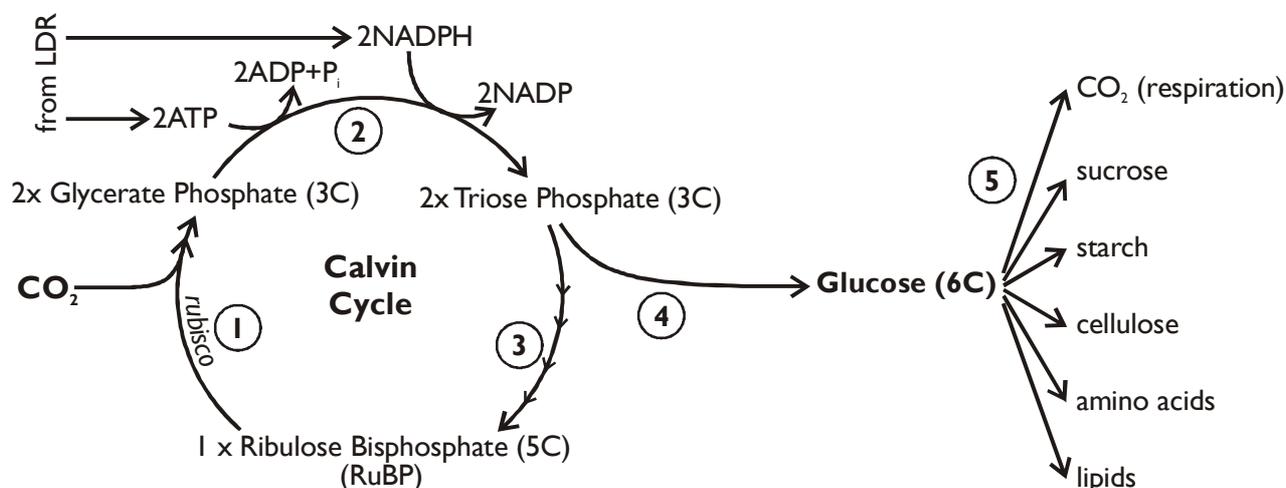
same way as respiration. This synthesis of ATP is called photophosphorylation because it uses light energy to phosphorylate ADP ($\text{ADP} + \text{P}_i \rightarrow \text{ATP}$).

Summary of the Light-Dependent Reactions

1. Light energy is absorbed by chlorophyll and used to photolyse water ($\text{H}_2\text{O} \rightarrow \text{O}_2 + \text{H}^+ + \text{e}^-$).
2. The high-energy electron is passed along the chain of proteins in the thylakoid membrane, gaining energy from light as it goes.
3. The electron is taken up by NADP, which is reduced to NADPH ($\text{NADP} + \text{H}^+ + \text{e}^- \rightarrow \text{NADPH}$).
4. The energy from the light is used to make ATP in the ATP synthase enzyme ($\text{ADP} + \text{P}_i \rightarrow \text{ATP}$).

The Light-Independent Reactions

The light-independent, or carbon-fixing reactions, of photosynthesis take place in the stroma of the chloroplasts and comprise another cyclic pathway, called the Calvin Cycle, after the American scientist who discovered it.

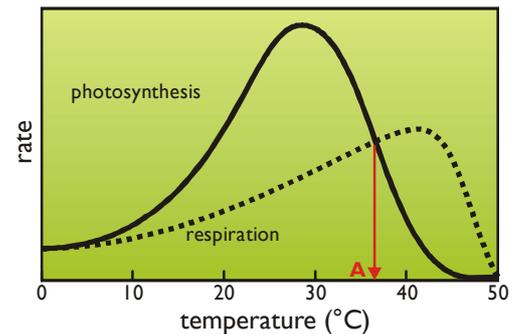


1. Carbon dioxide binds to the 5-carbon sugar ribulose biphosphate (RuBP) to form 2 molecules of the 3-carbon compound glycerate phosphate. This carbon-fixing reaction is catalysed by the enzyme ribulose biphosphate carboxylase, always known as rubisco. It is a very slow and inefficient enzyme, so large amounts of it are needed (recall that increasing enzyme concentration increases reaction rate), and it comprises about 50% of the mass of chloroplasts, making it the most abundant protein in nature.
2. Glycerate phosphate ($\text{C}_3\text{H}_4\text{O}_4\cdot\text{PO}_3$) is an acid, not a carbohydrate, so it is reduced and activated to form triose phosphate ($\text{C}_3\text{H}_6\text{O}_3\cdot\text{PO}_3$), the same 3-carbon sugar as that found in glycolysis. Two ATP and two NADPH molecules from the light-dependent reactions provide the energy for this step. The ADP and NADP return to the thylakoid membrane for recycling.
3. Triose phosphate is a branching point. Most of the triose phosphate continues through a complex series of reactions to regenerate the RuBP and complete the cycle. 5 triose phosphate molecules ($5 \times 3\text{C} = 15$ carbon atoms) combine to form 3 RuBP molecules ($3 \times 5\text{C} = 15$ carbon atoms).
4. Every 3 turns on average of the Calvin Cycle 3 CO_2 molecules are fixed to make 1 new triose phosphate molecule ($3\text{CO}_2 + 6\text{H} \rightarrow \text{C}_3\text{H}_6\text{O}_3$). This triose phosphate leaves the cycle, and two of these triose phosphate molecules combine to form one glucose molecule using the glycolysis enzymes in reverse.
5. The light-independent reactions are now finished, and the glucose can now be transported out of the chloroplast and used to make all the other organic compounds that the plant needs (cellulose, lipids, proteins, nucleic acids, etc). Some of these need the addition of mineral elements like N, P or S. Plants are very self-sufficient!

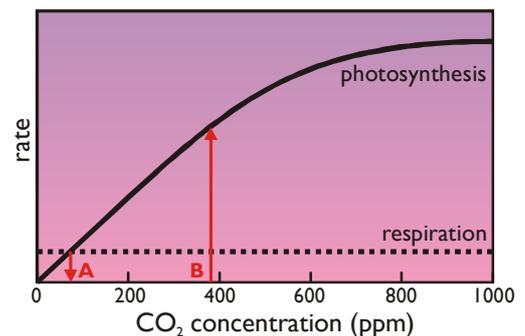
Factors affecting the rate of Photosynthesis

The rate of photosynthesis by a plant or alga can be measured by recording the amount of oxygen produced, or carbon dioxide used, in a given period of time. But these measurements are also affected by respiration, which plants do all the time, so the respiration rate must be measured separately. We've already seen that a plant's growth (or productivity) depends on the difference between the rates of photosynthesis and respiration (p 36). The conditions at which the rates of photosynthesis and respiration are equal, so there is no net change in oxygen or carbon dioxide concentration, is called the compensation point. Many of the environmental factors that affect photosynthesis also affect respiration.

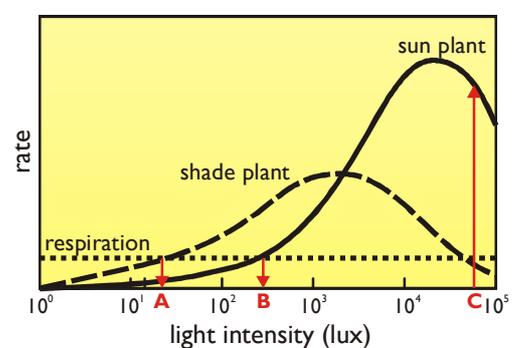
- **Temperature.** Temperature affects the rates of all enzyme reactions, so the rates of photosynthesis and respiration are both affected. Photosynthesis is more sensitive to temperature with an optimum of about 30-35°C, whereas respiration often has an optimum nearer to 45°C. So there is a temperature compensation point around 40°C (A), and above this temperature plants lose mass as the rate of respiration is greater than the rate of photosynthesis.



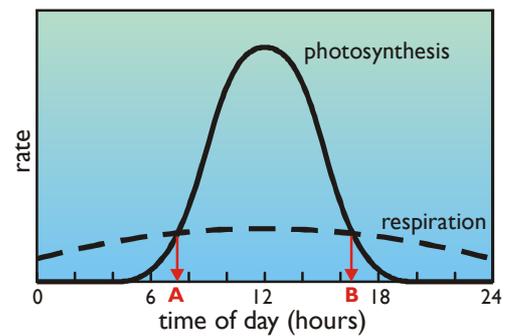
- **Carbon dioxide concentration.** Carbon dioxide is the substrate for the enzyme rubisco in the light-independent stages of photosynthesis, so the higher the carbon dioxide concentration the faster the rate of the Calvin cycle. The rate of respiration is not affected by carbon dioxide concentration, and the carbon dioxide compensation point is usually very low, at about 50ppm (A). Normal carbon dioxide concentration in the air is about 400ppm (B), whereas the optimum concentration for most plants is nearer to 1000ppm, so carbon dioxide is often the limiting factor.



- **Light intensity.** Light is the source of energy for the production of ATP and NADPH in the light-dependent stages of photosynthesis, so the higher the light intensity the faster the rate of photosynthesis. The rate of respiration is not affected by light intensity, and the light compensation point is usually low. Shade plants are adapted to growing in low light conditions (such as a forest floor), so have a very low light compensation point (A) and a low optimum intensity. Shade plants make good house plants, since they are adapted to the low light intensities indoors. Sun plants have a higher compensation point (B), and have a very high optimum near the light intensity of a bright summer's day (C).



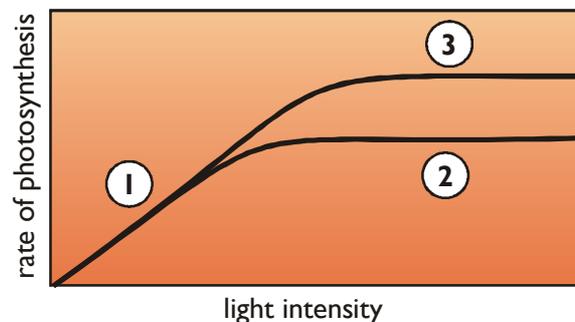
- Time of Day.** Both photosynthesis and respiration are affected by time of day: photosynthesis by changes in light and respiration by changes in temperature. At night respiration exceeds photosynthesis, while during the day photosynthesis exceeds respiration, so there are two compensation points each day (A and B). Over a 24-hour period the amount of photosynthesis is greater than the amount of respiration, so plants gain mass and have a net uptake of carbon dioxide.



Understanding how factors affect photosynthesis and respiration is very important for farmers and commercial growers. For example in a closed greenhouse with lots of plants the carbon dioxide concentration can fall very low, so it can be worth increasing the CO₂ concentration in the greenhouse to increase the rate of photosynthesis. This is most efficiently done by burning a fuel, since this releases CO₂ and raises the temperature. It's hard to beat the intensity of daylight, but day length can be increased with artificial lighting.

Limiting Factors

Although all these factors (and many others) could affect the rate of photosynthesis, at any given time there can only be **one** factor that is actually controlling the rate – the **limiting factor**. This is the factor that is in shortest supply, or furthest from its optimum. It's a bit like a chain that is only as strong as its weakest link. We can observe this in an experiment to investigate the rate of photosynthesis.



- At low light intensities the rate of photosynthesis increases as the light intensity increases. This must mean that light is the limiting factor, since the rate depends on it.
- At higher light intensities the rate of photosynthesis stays the same even if the light intensity increases. This means that light is not the limiting factor, since the rate doesn't depend on it. This isn't surprising since there is now plenty of light. The rate of photosynthesis must be limited by some other factor.
- If we repeat the experiment at a higher concentration of carbon dioxide we get a higher rate, showing that a higher rate is possible, and that carbon dioxide concentration was rate limiting at point 2.

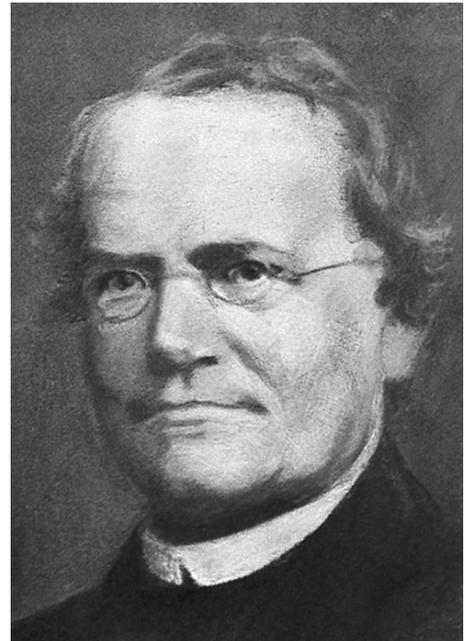
Mendelian Genetics

In unit 2 we studied molecular genetics – the study of DNA. Here we are concerned with transmission genetics, which is the study of inheritance of characteristics at the whole organism level. This is also known as classical genetics or Mendelian genetics, since it was pioneered by Gregor Mendel.

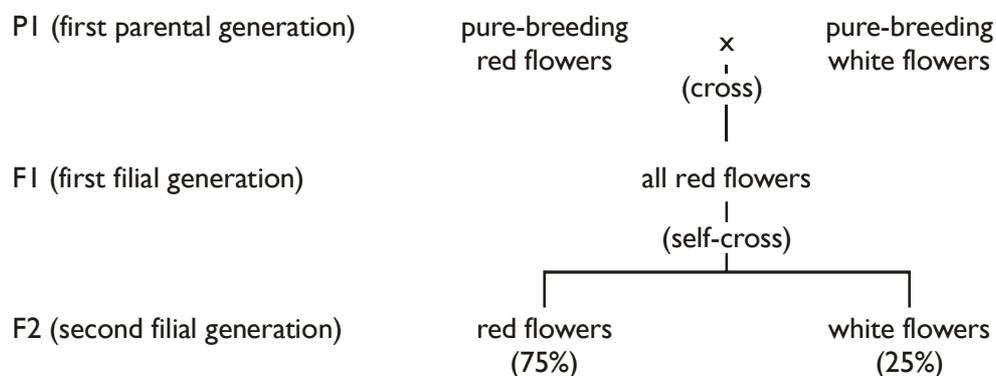
Gregor Mendel

Mendel (1822-1884) was an Austrian monk at Brno monastery. He was a keen scientist and gardener, and studied at Vienna University, where he learnt mathematics. He investigated inheritance in pea plants and published his results in 1866. They were ignored at the time, but were rediscovered in 1900, and Mendel is now recognised as the “Father of Genetics”. His experiments succeeded where other had failed because:

- Mendel investigated simple qualitative characteristics (or traits), such as flower colour or seed shape, and he varied one trait at a time. Previous investigators had tried to study many complex quantitative traits, such as human height or intelligence, but this is a rare instance where qualitative results are more informative than quantitative ones, and Mendel knew this.
- Mendel used an organism whose sexual reproduction he could easily control by carefully pollinating stigmas with pollen using a brush. Peas can also be self pollinated, allowing self crosses to be performed. This is not possible with animals.
- Mendel repeated his crosses hundreds of times and applied statistical tests to his results.
- Mendel studied two generations of peas at a time.



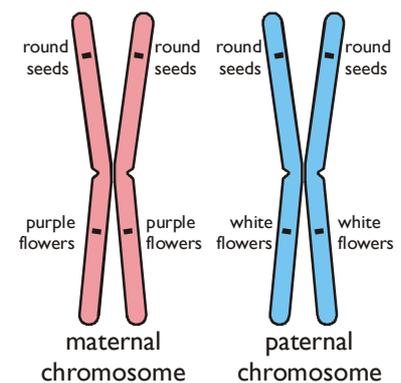
A typical experiment looked like this:



Mendel made several conclusions from these experiments:

1. There are no mixed colours (e.g. pink), so this disproved the widely-held blending theories of inheritance that characteristics gradually mixed over time.
2. A characteristic can disappear for a generation, but then reappear the following generation, looking exactly the same. So a characteristic can be present but hidden.
3. The outward appearance (the phenotype) is not necessarily the same as the inherited factors (the genotype) For example the P1 red plants are not the same as the F1 red plants.
4. One form of a characteristic can mask the other. The two forms are called dominant and recessive respectively.
5. The F2 ratio is always close to 3:1 (or 75%:25%). Mendel was able to explain this by supposing that each individual has two versions of each inherited factor, one received from each parent. We'll look at his logic in a minute.

Mendel's factors are now called genes and we know they are found on chromosomes. The two alternative forms are called alleles and are found on homologous pairs of chromosomes (the maternal and paternal). So in the example above we would say that there is a gene for flower colour and its two alleles are "red" and "white". One allele comes from each parent, and the two alleles are found on the same position (or locus) on the homologous chromosomes. If the homologous chromosomes have the same alleles at a locus this is homozygous, and if they have different alleles this is heterozygous. The chromosomes on the right are homozygous for the seed shape genes but heterozygous for the flower colour gene. The term "pure-breeding" really means homozygous. You should revise genes and chromosomes from unit 2.



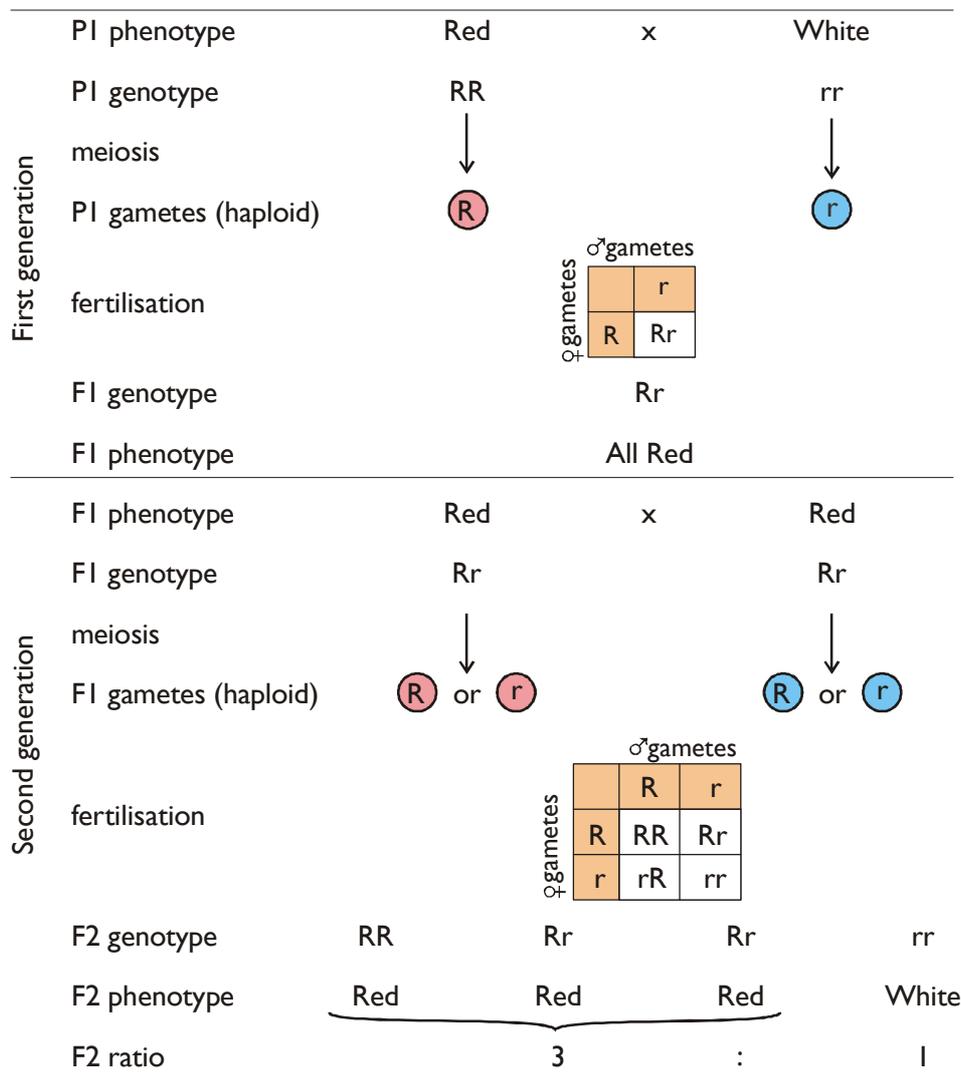
With two alleles there are three possible combinations of alleles (or genotypes) and two possible appearances (or phenotypes):

Genotype	Name	Phenotype
RR	homozygous dominant	red
rr	homozygous recessive	white
Rr, rR	heterozygous	red

The dominant allele is defined as the allele that is expressed in the heterozygous state, while the recessive allele is defined as the allele that is only expressed in the homozygous state (or is not expressed in the heterozygous state).

The Monohybrid Cross

A simple breeding experiment involving just a single characteristic, like Mendel's experiment, is called a monohybrid cross. We can now explain Mendel's monohybrid cross in detail.



At fertilisation any male gamete can fertilise any female gamete at random. The possible results of a fertilisation can most easily be worked out using a Punnett Square as shown in the diagram. Each of the possible outcomes has an equal chance of happening, so this explains the 3:1 ratio observed by Mendel.

This is summarised in Mendel's First Law, which states that individuals carry two discrete hereditary factors (alleles) controlling each characteristic. The two alleles segregate (or separate) during meiosis, so each gamete carries only one of the two alleles.

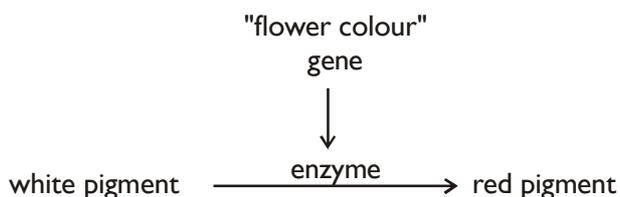
The Test Cross

You can see an individual's phenotype, but you can't see its genotype. If an individual shows the recessive trait (white flowers in the above example) then they must be homozygous recessive as it's the only genotype that will give that phenotype. If they show the dominant trait then they could be homozygous dominant or heterozygous. You can find out which by performing a test cross with a pure-breeding homozygous recessive. This gives two possible results:

- If the offspring all show the dominant trait then the parent must be homozygous dominant.
- If the offspring are a mixture of phenotypes in a 1:1 ratio, then the parent must be heterozygous.

How does Genotype control Phenotype?

Mendel never knew this, but we can explain in detail the relation between an individual's genes and its appearance. A gene was originally defined as an inherited factor that controls a characteristic, but we now know that a gene is also a length of DNA that codes for a protein (see unit 2). It is the proteins that actually control phenotype in their many roles as enzymes, pumps, transporters, motors, hormones, or structural elements. For example the flower colour gene actually codes for an enzyme that converts a white pigment into a red pigment:



- The dominant allele is the normal (or “wild-type”) form of the gene that codes for functioning enzyme, which therefore makes red-coloured flowers.
- The recessive allele is a mutation of the gene. This mutated gene codes for non-functional enzyme, so the red pigment can't be made, and the flower remains white. Almost any mutation in a gene will result in an inactive gene product (often an enzyme), since there are far more ways of making an inactive protein than a working one.

Sometimes the gene actually codes for a protein apparently unrelated to the phenotype. For example the gene for seed shape in peas (round or wrinkled) actually codes for an enzyme that synthesises starch! The functional enzyme makes lots of starch and the seeds are full and rounded, while the non-functional enzyme makes less starch so the seeds wrinkle up. The gene responsible for all the symptoms of cystic fibrosis actually codes for a chloride ion channel protein. A “tallness” gene may be a control gene that regulates the release of growth hormone.

This table shows why the allele that codes for a functional protein is usually dominant over an allele that codes for a non-function protein. In a heterozygous cell, some functional protein will be made, and this is usually enough to have the desired effect. In particular, enzyme reactions are not usually limited by the amount of enzyme, so a smaller amount in heterozygotes will have little effect on phenotype.

Genotype	Gene product	Phenotype
homozygous dominant (RR)	all functional enzyme	red
homozygous recessive (rr)	no functional enzyme	white
heterozygous (Rr)	some functional enzyme	red

Sex Determination

In unit 2 we came across the sex chromosomes (X and Y). Since these are non-homologous they are called heterosomes, while the other 22 pairs are called autosomes. In humans the sex chromosomes are homologous in females (XX) and non-homologous in males (XY), though in other species it is the other way round. The inheritance of the X and Y chromosomes can be demonstrated using a monohybrid cross:

P1 phenotype	female	x	male						
P1 genotype	XX		XY						
P1 gametes	⊗		⊗ or ⊙						
fertilisation		♂gametes							
		<table border="1" style="border-collapse: collapse; text-align: center;"> <tr> <td style="padding: 2px 5px;">♀gametes</td> <td style="padding: 2px 5px;">X</td> <td style="padding: 2px 5px;">Y</td> </tr> <tr> <td style="padding: 2px 5px;">X</td> <td style="padding: 2px 5px;">XX</td> <td style="padding: 2px 5px;">XY</td> </tr> </table>	♀gametes	X	Y	X	XX	XY	
♀gametes	X	Y							
X	XX	XY							
F1 genotype	XX		XY						
F1 phenotype	female		male						

This shows that there will always be a 1:1 ratio of males to females. Note that female gametes (eggs) always contain a single X chromosome, while the male gametes (sperm) can contain a single X or a single Y chromosome. Sex is therefore determined solely by the sperm. There are techniques for separating X and Y sperm, and this is used for planned sex determination in farm animals using artificial insemination (AI).

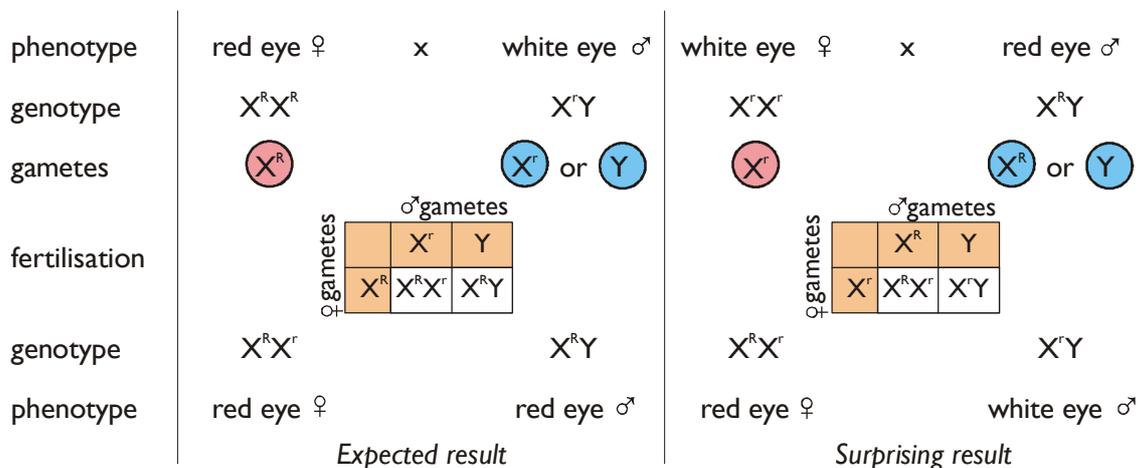
In humans it is the Y chromosome that actually determines sex: all embryos start developing as females, but if the sex-determining “SRY” gene on the Y chromosome is expressed, male hormones are produced in the embryo, causing the development of male characteristics. In the absence of male hormones, the embryo continues to develop as a female. The X chromosome is not involved in sex determination.

Sex-Linked Characteristics

What else do the X and Y chromosomes do? As we saw in unit 2, the Y chromosome is very small, containing very few genes, and doesn't seem to do much other than determine sex. The X chromosome, on the other hand, is large and contains over a thousand genes that have nothing to do with sex, coding for important products such as rhodopsin, blood clotting proteins and muscle proteins. Females have two copies of each gene on the X chromosome (i.e. they're diploid), but males only have one copy of each gene on the X chromosome (i.e. they're haploid). This means that the inheritance of these genes is different for males and females, so they are called sex linked characteristics.

Eye Colour in Fruit Flies

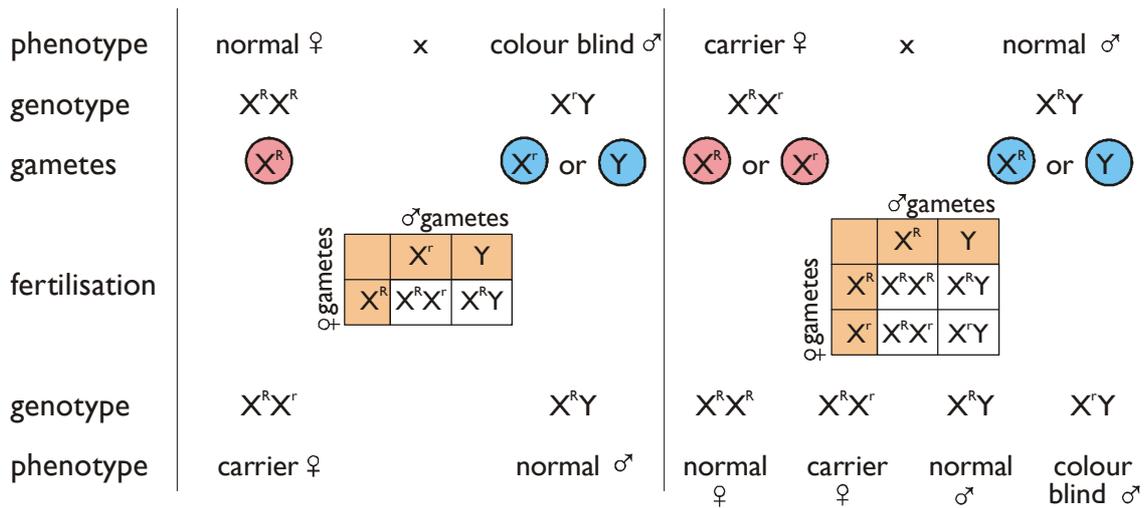
The first example of sex linked genes discovered was eye colour in *Drosophila* fruit flies. Red eyes (R) are dominant to white eyes (r) and when a red-eyed female is crossed with a white-eyed male, the offspring all have red eyes, as expected for a dominant characteristic (left cross below). However, when the opposite cross was done (a white-eye male with a red-eyed male) all the male offspring had white eyes (right cross below). This surprising result was not expected for a simple dominant characteristic, but it could be explained if the gene for eye colour was located on the X chromosome. Note that in these crosses the alleles are written in the form X^R (red eyes) and X^r (white eyes) to show that they are on the X chromosome.



Males always inherit their X chromosome from their mothers, and always pass on their X chromosome to their daughters.

Colour Blindness

Another well-known example of a sex linked characteristic is colour blindness in humans. 8% of males are colour blind, but only 0.7% of females. There are three different light-receptor proteins involved in colour vision, sensitive to red, green and blue light. The genes for the green-sensitive and red-sensitive proteins are on the X chromosome, and mutations in either of these lead to colour blindness. The diagram below shows two crosses involving colour blindness, using the symbols X^R for the dominant allele (normal rhodopsin, normal vision) and X^r for the recessive allele (non-functional rhodopsin, colour blind vision).



Other examples of sex linked characteristics include haemophilia and muscular dystrophy.

Codominance

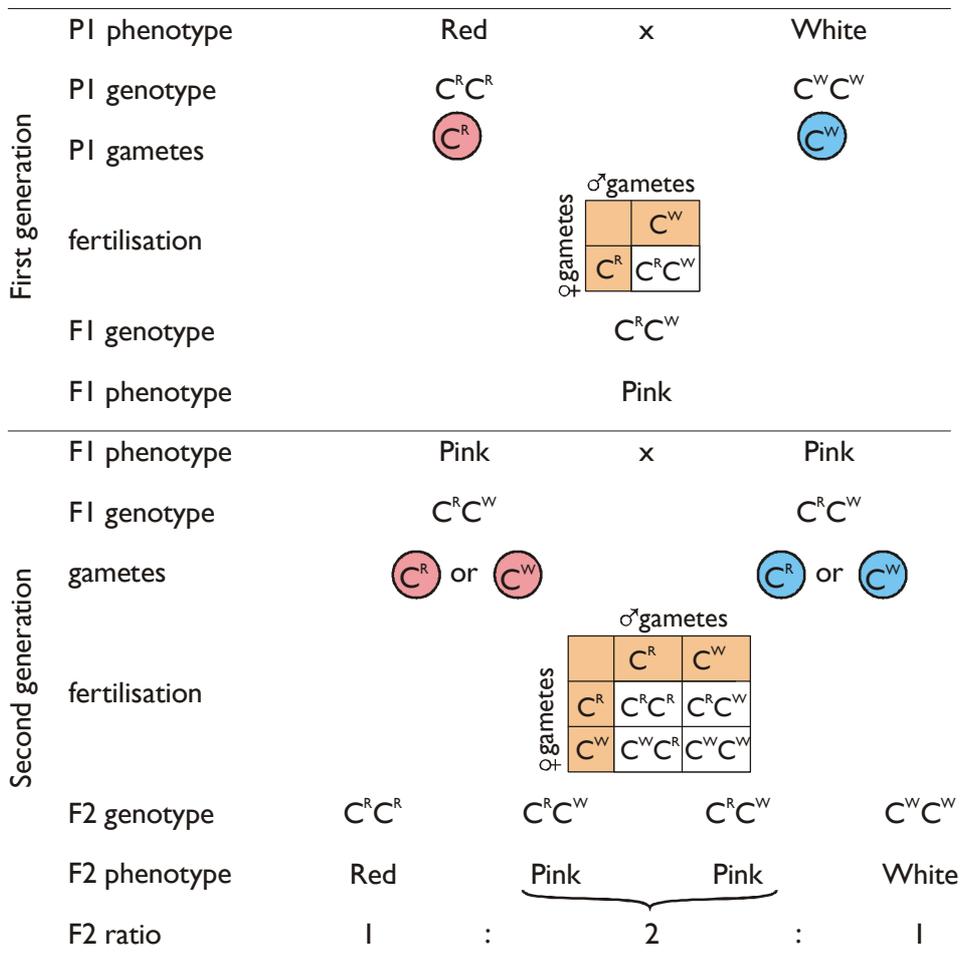
In most situations (and all of Mendel's experiments) one allele is completely dominant over the other, so there are just two phenotypes. But in some cases there are three phenotypes, because neither allele is completely dominant over the other, so the heterozygous genotype has its own phenotype. This situation is called codominance or incomplete dominance. Since there is no dominance we can no longer use capital and small letters to indicate the alleles, so a more formal system is used. The gene is represented by a letter and the different alleles by superscripts to the gene letter.

Flower Colour in Snapdragons

One example of codominance is flower colour in snapdragon plants. The flower colour gene C has two alleles: C^R (red) and C^W (white). The three genotypes and their phenotypes are:

Genotype	Gene product	Phenotype
homozygous (C ^R C ^R)	all functional enzyme	red
Homozygous (C ^W C ^W)	no functional enzyme	white
heterozygous (C ^R C ^W)	some functional enzyme	pink

In this case the enzyme is probably less active, so a smaller amount of enzyme will make significantly less product, and this leads to the third phenotype. A monohybrid cross looks like this:



Note that codominance is not an example of “blending inheritance” since the original phenotypes reappear in the second generation. The genotypes are not blended and they still obey Mendel’s law of segregation. It is only the phenotype that appears to blend in the heterozygotes.

Sickle Cell Anaemia

Another example of codominance is sickle cell haemoglobin in humans. The gene for haemoglobin (or more accurately for the polypeptide globin – see unit 1) “Hb” has two codominant alleles: Hb^A (the normal gene) and Hb^S (the mutated gene). The mutation in the Hb^S gene is a single base substitution (T→A), changing one amino acid out of 146 in the polypeptide chain. This amino acid binds to other haemoglobin molecules, so the molecules link together to form long chains, distorting the red blood cells into sickle shapes.



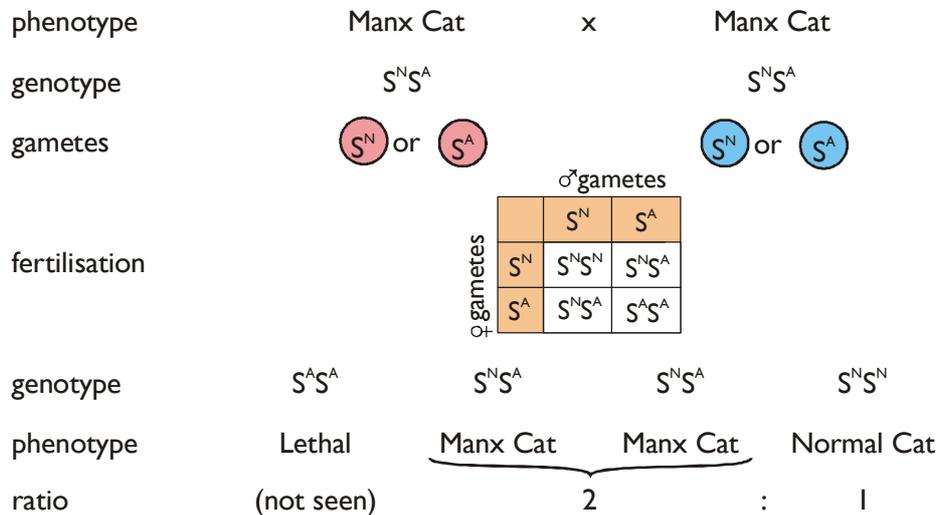
There are three phenotypes:

- Hb^AHb^A **Normal.** All haemoglobin molecules are normal, with normal disk-shaped red blood cells.
- Hb^SHb^S **Sickle cell anaemia.** All haemoglobin molecules are abnormal, so most red blood cells are sickle-shaped. These sickled red blood cells are less flexible than normal cells, so can block capillaries and arterioles, causing cell death and severe pain. Sickle cells are also destroyed by the spleen faster than they can be made, so not enough oxygen can be carried in the blood (anaemia). Without treatment this phenotype is fatal in early childhood, though modern medical intervention can extend life expectancy to 50.
- Hb^AHb^S **Sickle cell trait.** 50% of the haemoglobin molecules in every red blood cell are normal, and 50% abnormal. Long chains do not form, so the red blood cells are normal and carry oxygen normally. However these red blood cells do sickle when infected by the malaria parasite, so infected cells are destroyed by the spleen. This phenotype therefore confers immunity to malaria, and is common in areas of the world where malaria is endemic.

Other examples of codominance include coat colour in cattle (red/white/roan), and coat colour in cats (black/orange/tortoiseshell).

Lethal Alleles

An unusual effect of codominance is found in Manx cats, which have no tails. If two Manx cats are crossed the litter has ratio of 2 Manx kittens to 1 normal (long-tailed) kitten. The explanation for this unexpected ratio is explained in this genetic diagram:



The gene S actually controls the development of the embryo cat's spine. It has two codominant alleles: S^N (normal spine) and S^A (abnormal, short spine). The three phenotypes are:

$S^N S^N$ **Normal.** Normal spine, long tail

$S^N S^A$ **Manx Cat.** Last few vertebrae absent, so no tail.

$S^A S^A$ **Lethal.** Spine doesn't develop, so this genotype is fatal early in development. The embryo doesn't develop and is absorbed by the mother, so there is no evidence for its existence.

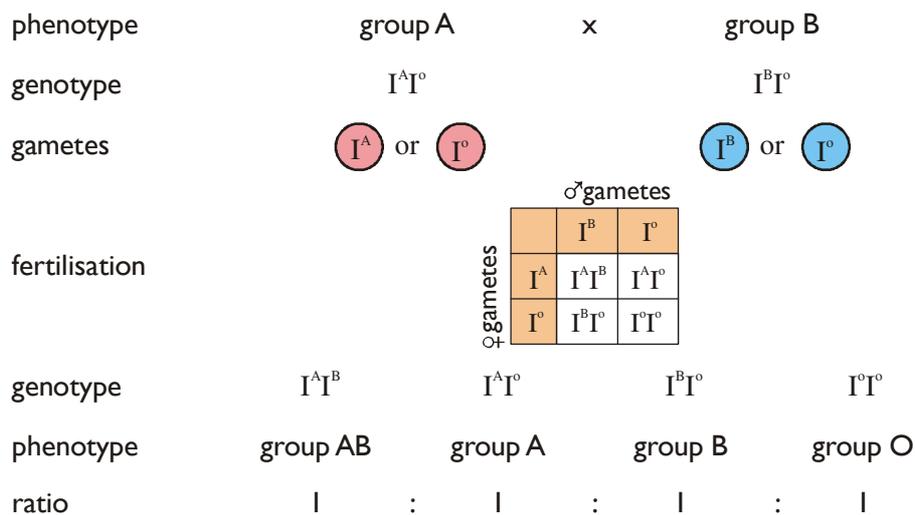
Many human genes also have lethal alleles, because many genes are so essential for life that a mutation in these genes is fatal. If the lethal allele is expressed early in embryo development then the fertilised egg may not develop enough to start a pregnancy, or the embryo may miscarry. If the lethal allele is expressed later in life, then we call it a genetic disease, such as muscular dystrophy or cystic fibrosis.

Multiple Alleles

An individual has two copies of each gene, so can only have two alleles of any gene, but there can be more than two alleles of a gene in a population. An example of this is blood group in humans. The red blood cell antigen is coded for by the gene I (for isohaemagglutinin), which has three alleles I^A , I^B and I^o . (They are written this way to show that they are alleles of the same gene.) I^A and I^B are codominant, while I^o is recessive. The six possible genotypes and four phenotypes are:

Phenotype (blood group)	Genotypes	antigens on red blood cells	plasma antibodies
A	$I^A I^A, I^A I^o$	A	anti-B
B	$I^B I^B, I^B I^o$	B	anti-A
AB	$I^A I^B$	A and B	none
O	$I^o I^o$	none	anti-A and anti-B

The cross below shows how all four blood groups can arise from a cross between a group A and a group B parent.



Other examples of multiple alleles are: eye colour in fruit flies, with over 100 alleles, and human leukocyte antigen (HLA) genes, with 47 known alleles.

Population Genetics and the Gene Pool

We've seen how alleles are passed on from one individual to another in a population. Now we'll see how all the alleles in a population might change. The sum of all the alleles of all the genes of all the individuals in a population is called the gene pool. We saw in unit 2 that genetic diversity in the gene pool was important for a population's survival.

The Hardy-Weinberg Principle

In the early 20th century biologists started to apply Mendel's laws of inheritance to whole populations. It was realised that the frequencies of dominant and recessive alleles would remain constant over time, so long as five key conditions about the population were met:

1. There are no mutations, so no new alleles are created.
2. There is no immigration, so no new alleles are introduced, and no emigration, so no alleles are lost.
3. There is no selection, so no alleles are favoured or eliminated.
4. Mating is random, so alleles are mixed randomly.
5. The population is large, so there are no genetic bottlenecks.

These conditions mean that there is nothing to disturb the gene pool, which therefore remains in a stable genetic equilibrium. In other words, the allele frequencies in the population will remain constant from generation to generation. This principle is called the Hardy-Weinberg principle, since it was devised independently by the English mathematician G. H. Hardy and the German physician G. Weinberg in 1908. Before this it was thought that dominant alleles would increase in frequency over time, and recessive alleles would decrease in frequency, but this intuitive idea is wrong. Dominant alleles need not be common. For example the dominant allele for Huntington's disease is very rare in the population and almost everyone is homozygous recessive.

The Hardy-Weinberg principle can be tested by measuring allele frequencies over time, and it is often found that the frequencies do change. This means that the at least one of the five conditions is not true, and the gene pool is not stable. In other words the population is evolving. So the Hardy-Weinberg principle provides a means of detecting evolution, and quantifying the rate of evolutionary change.

The Hardy-Weinberg Equation

If the gene pool is stable then we can use a simple equation to calculate the gene frequencies in a population. This is called the Hardy-Weinberg equation. There are three kinds of frequencies:

- **Phenotype frequencies** are proportions of the different characteristics in the population (e.g. red or white). These are the easiest, because we can see and count them in a population.
- **Genotype frequencies** are the proportions of the three possible genotypes (BB, Bb and bb) in the population. This isn't so easy, because we can't see the genotypes, but we can calculate them.
- **Allele frequencies** are the proportions of the two alleles B and b in the population. Allele frequencies are particularly interesting because evolution causes the allele frequencies to change.

Just as with the genetic crosses, let's consider the case of a single gene at a time. For example, imagine that coat colour in cats is controlled by a single gene with two alleles – black (B) and white (b). The black allele is completely dominant over the white allele. Each cat has two alleles for coat colour – either BB or Bb or bb. In population genetics we always measure frequencies – decimal fractions out of one. We don't know what the frequency of each genotype in the population is, but we do know that the sum of the two allele frequencies must add up to one, by definition (because there are only two alleles of this gene). Mathematically, if p is the frequency of the dominant allele A, and q is the frequency of the recessive allele a, then

$$p + q = 1$$

Now the gametes produced by the cats in this population will only have one allele of the coat colour gene each – either B or b. While we don't know the allele in any particular gamete, we know that overall, because gamete production is random, the frequencies of the B and b alleles in the gametes will be the same as in the gene pool of the parent cats, i.e. p and q . So we can do a Punnett square for reproduction in this cat population:

		♂ gametes	
		B (frequency p)	b (frequency q)
♀ gametes	B (frequency p)	BB (frequency p^2)	Bb (frequency pq)
	b (frequency q)	Bb (frequency pq)	bb (frequency q^2)

This Punnett square gives us the frequencies of the different genotypes in the population when the cats reproduce. The genotype BB has a frequency p^2 , the genotype bb has a frequency q^2 , and the genotype Bb has a frequency $2pq$. The sum of the genotype frequencies must add up to one (by definition), so:

$$p^2 + 2pq + q^2 = 1$$

This is the Hardy-Weinberg equation,

Using the Hardy-Weinberg Equation

We can use the Hardy-Weinberg equation to calculate genotype and allele frequencies from observed phenotype frequencies. Let's take a population of 1000 cats with 840 black cats and 160 white cats.

⇒ The phenotype frequency for black is 0.84 (840/1000) and for white is 0.16 (160/1000)

⇒ We know that white is the recessive allele, so the white cats must be homozygous recessive, so the frequency of the genotype bb is 0.16

⇒ The genotype bb has a frequency q^2 , so $q^2 = 0.16$

⇒ $q = \sqrt{q^2} = \sqrt{0.16} = 0.4$

⇒ $p + q = 1$, so $p = 1 - q = 1 - 0.4 = 0.6$

Now we can calculate the genotype frequencies:

⇒ frequency of $BB = p^2 = 0.6^2 = 0.36$

⇒ frequency of $Bb = 2pq = 2 \times 0.6 \times 0.4 = 0.48$

⇒ frequency of $bb = q^2 = 0.16$ (already found)

⇒ check that these add up to one = 1.00

We can convert these frequencies to actual numbers in the population, for example

⇒ Number of heterozygous cats = $0.48 \times 1000 = 480$

The Hardy-Weinberg equation can be used to calculate any of the three types of frequencies:

- **Allele frequencies.** The frequency of the recessive allele (a) is q and the frequency of the dominant allele (A) is p .
- **Genotype frequencies.** The frequency of the homozygous recessive genotype (aa) is q^2 ; the frequency of the homozygous dominant genotype (AA) is p^2 ; and the frequency of the heterozygous genotype (Aa) is $2pq$.
- **Phenotype frequencies.** The frequency of the recessive phenotype is q^2 , and the frequency of the dominant phenotype is $p^2 + 2pq$.

The Hardy-Weinberg equation can be very useful in many different applications. For example the incidence of the single-gene recessive disorder cystic fibrosis in humans is 1 in 2500. From this observation we can use the Hardy-Weinberg equation to calculate that one in 25 people are heterozygous carriers of the disease allele, and this sort of information is important in genetic counselling.

Evolution and Natural Selection

History of Ideas of Life on Earth

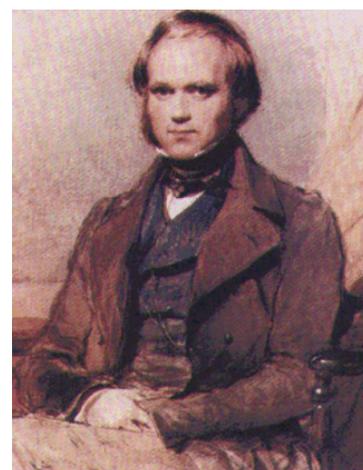
17th Century Most people believed in Creationism, which considered that all life was created just as it is now. This was not based on any evidence, but was instead a belief.

18th Century Naturalists began systematic classification systems and noticed that groups of living things had similar characteristics and appeared to be related. So their classifications looked a bit like family trees.

European naturalists travelled more widely and discovered more fossils, which clearly showed that living things had changed over time, so were not always the same. Extinctions were also observed (e.g. dodo), so species were not fixed. Selective breeding was widely practised and it was realised that species (like crops, working dogs, racing pigeons) could be changed dramatically by selection.

19th Century Lamarck (1809) proposed a hypothesis that living things changed by inheriting acquired characteristics. e.g. giraffes stretched their necks to reach food, and their offspring inherited stretched necks. This is now known to be wrong, since many experiments (and experience) have shown that acquired characteristics are not inherited. Nevertheless Lamarck's theory was one of the first to admit that species changed, and to try to explain the change.

Charles Darwin (1859) published "*On the origin of species by means of natural selection, or the preservation of favoured races in the struggle for life*", which proposed the idea of natural selection, a far better explanation for the changes in species. "Origin" has since been recognised as one of the most important books ever written. A very similar theory was also proposed by Alfred Wallace, and Darwin and Wallace agreed to publish at the same time.



20th Century Mendel's work on genetics was rediscovered and combined with Darwin's theory to form modern Darwinism. Many new techniques, like fossil dating, molecular biology, microbiology and mathematical modelling, gradually formed an extensive and overwhelming body of experimental evidence for Darwinism.

Darwin's Theory of Evolution by Natural Selection

Darwin's theory was based on four observations:

- Individuals within a species differ from each other – there is variation.
- Offspring resemble their parents – characteristics are inherited.
- Far more offspring are generally produced than survive to maturity – most organisms die young from predation, disease and competition.
- Populations are usually fairly constant in size.

Darwin realised that the organisms that die young were not random, but were selected by their characteristics. He concluded that individuals that were better adapted to their environment compete better than the others, survive longer and reproduce more, so passing on more of their successful genes to the next generation.

Darwin explained the giraffe's long neck as follows:

1. In a population of horse-like animals there would be random genetic variation in neck length.
2. In an environment where there were trees and bushes, the longer-necked animals were slightly better adapted as they could reach more leaves, and so competed well compared to their shorter-necked relatives. These longer-necked animals lived longer, through more breeding seasons, and so had more offspring.
3. The shorter-necked animals would be more likely to lose the competition for food, so would be poorly nourished and would probably die young from predation or disease. They would have few, if any, offspring.
4. So in the next generation there were more long-neck alleles than short-neck alleles in the population. If this continued over very many generations, then in time the frequency of long-neck alleles would increase and so the average neck length would increase.

[Today it is thought more likely that the selection was for long legs to run away from predators faster, and if you have long legs you need a long neck to be able to drink. But the process of selection is just the same.]

Darwin wasn't the first to suggest evolution of species, but he was the first to suggest a plausible mechanism for the evolution - natural selection, and to provide a wealth of evidence for it.

Darwin used the analogy of selective breeding (or artificial selection) to explain natural selection. In selective breeding, desirable characteristics are chosen by humans, and only those individuals with the best characteristics are used for breeding. In this way species can be changed over a period of time. All domesticated species of animal and plant have been selectively bred like this, often for thousands of years, so that most of the animals and plants we are most familiar with are not really natural and are nothing like

their wild relatives (if any exist). The analogy between artificial and natural selection is a very good one, but there is one important difference - Humans have a goal in mind; nature does not.

Summary of Natural Selection

1. There is genetic variation in a characteristics within a population
2. Individuals with characteristics that make them less well adapted to their environment will die young from predation, disease or competition.
3. Individuals with characteristics that make them well adapted to their environment will survive and reproduce.
4. The allele frequency will change in each generation.

Examples of Natural Selection

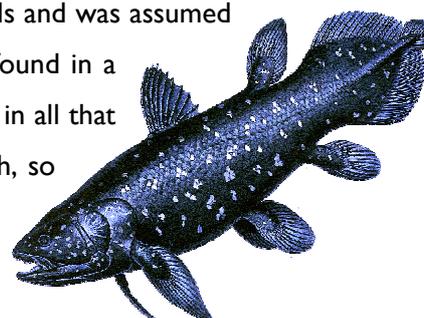
We'll look at some examples of natural selection in action. In fact most things you've studied in the biology course (like protein structure, lung anatomy, the nitrogen cycle, disease, anything) are examples of natural selection. It has been said that nothing in biology makes sense, except in the light of evolution.

The Peppered Moth. These light-coloured moths are well camouflaged from bird predators against pale lichen-covered bark of trees, while rare mutant dark moths are easily picked off. During the industrial revolution in the 19th century, birch woods near industrial centres became black with pollution. In this changed environment the black moths had a selective advantage and became the most common colour, while the pale moths were easily predated and became rare. Kettlewell tested this by releasing dark and light moths in polluted and unpolluted environments and observing selective predation. Since pollution has cleared up in the 20th century the selection has reversed again and pale moths are now favoured again over dark ones.



Bacterial resistance to antibiotics. Antibiotics kill bacteria, but occasionally a chance mutant bacterium appears that is resistant to an antibiotic. In an environment where the antibiotic is often present, this mutant has an enormous selective advantage since all the normal (wild type) bacteria are killed leaving the mutant cell free to reproduce and colonise the whole environment without any competition. Some farmers routinely feed antibiotics to their animals to prevent infection, but this is a perfect environment for resistant bacteria to thrive. The best solution is to stop using the antibiotic so that the resistant strain has no selective advantage, and may die out.

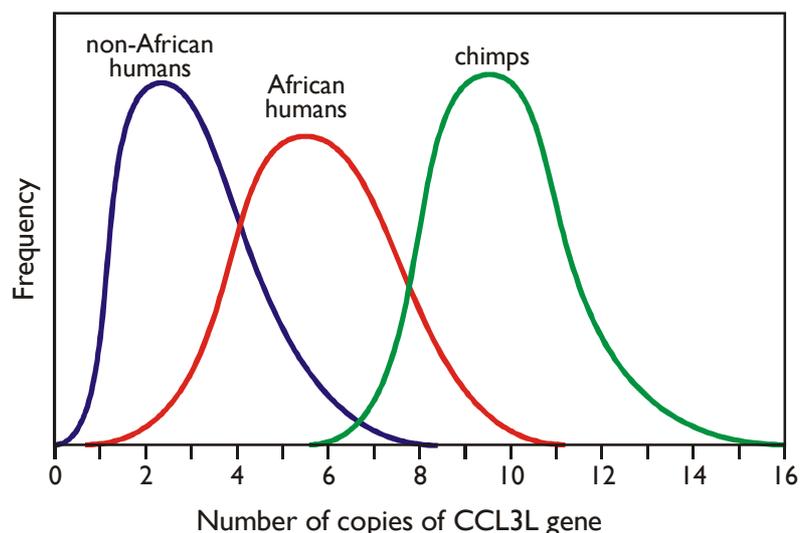
The Coelocanth. This fish species was known only from ancient fossils and was assumed to have been extinct for 70 million years until a living specimen was found in a trawler net off South Africa in 1938. So this species has hardly changed in all that time. The deep ocean is one of the most stable environments on Earth, so there was little selection pressure to change.



Lactose tolerance in humans. Some people are lactose intolerant, and feel ill (including diarrhoea and vomiting) when they drink milk. In fact globally most human adults are lactose intolerant and this is the normal condition: lactose tolerance in adults is a mutation. All infant mammals make lactase to digest lactose in their mother's milk, and they all stop producing lactase after they are weaned (its production is switched off at about age four in most humans).

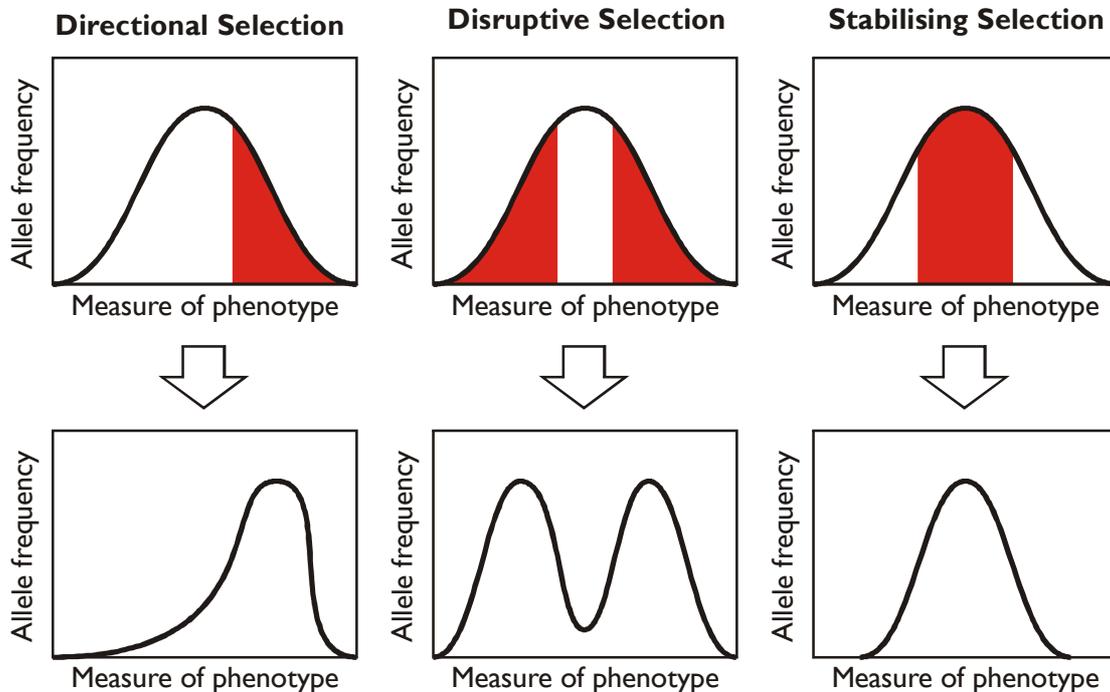
Around 10,000 years ago humans gradually changed from being mainly hunter-gatherers to being mainly farmers, and for the first time animal milk was available as a food source. Humans who, through a chance mutation, could drink milk without feeling ill were at an advantage, as they could supplement their normal diet with milk in harsh times (and farming was very unreliable in the early days). By natural selection they survived and their genes spread in their populations. As a result in human societies that adopted pastoral (animal) farming (such as most Europeans, northern Indians and some Africans) are generally lactose tolerant today, while the rest (most Asians, Africans, native Americans and Australians) remain lactose intolerant as adults.

HIV resistance in humans. The AIDS virus HIV first arose in human populations in the 1930s in West Africa, where it spread from primates through the practice of killing and eating "bush meat". Since then it has gradually spread around the world. Why is HIV so fatal to humans, but has so little effect on chimps? It turns out that chimps are resistant because they have a protein (called CCL3) that stops HIV entering and infecting white blood cells. Now some humans have this protein too, and it seems that the more copies of the gene for CCL3 you have, the more resistant you are to HIV. Chimps have on average 11 copies of the CCL3 gene, African humans have on average 6 copies, and non-African humans have on average 2 copies. In Africa people who, by chance, have many copies are favoured and will reproduce, while those with few copies die young without reproducing. So natural selection in humans explains the frequency of the CCL3 gene. A thousand years in the future, if we have not developed a medical cure for HIV, the whole human population will probably have evolved to possess around 11 copies of CCL3.



Types of Natural Selection

Populations change over time as their environment changes. These changes can be recorded as changing histograms of a particular phenotype (which of course is due to changes in the underlying alleles). These histograms show three kinds of natural selection, depending on which phenotypes are selected by the environment. The shaded areas represent the phenotypes that are favoured.



- **Directional Selection** occurs when one extreme phenotype (e.g. tallest) is favoured over the other extreme (e.g. shortest). This happens when the environment changes in a particular way. "Environment" includes biotic as well as abiotic factors, so organisms evolve in response to each other. e.g. if predators run faster there is selective pressure for prey to run faster, or if one tree species grows taller, there is selective pressure for other to grow tall. Most environments do change (e.g. due to migration of new species, or natural catastrophes, or climate change, or to sea level change, or continental drift, etc.), so directional selection is common.
- **Disruptive (or Diverging) Selection.** This occurs when both extremes of phenotype are selected over intermediate types. For example in a population of finches, birds with large and small beaks feed on large and small seeds respectively and both do well, but birds with intermediate beaks have no advantage, and are selected against.
- **Stabilising (or Normalising) Selection.** This occurs when the intermediate phenotype is selected over extreme phenotypes, and tends to occur when the environment doesn't change much. For example birds' eggs and human babies of intermediate birth weight are most likely to survive. Natural selection doesn't have to cause a directional change, and if an environment doesn't change there is no pressure for a well-adapted species to change. Fossils suggest that many species remain unchanged for long periods of geological time.

The Origin of New Species – Speciation

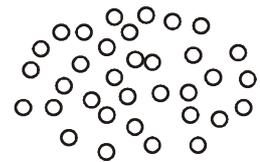
The examples of evolution by natural selection we have just seen don't always give rise to new species, though they do illustrate change. But the 100 million species that do and have existed arose by evolution, so we need to understand how.

In unit 2 we defined a species as:

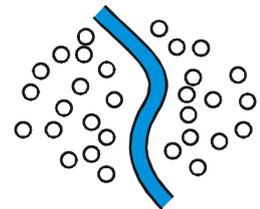
- Organisms in the same species are similar in appearance (morphology), behaviour and biochemistry, and have the same ecological niche.
- Organisms in the same species can breed together in their natural environment to produce fertile offspring, but cannot breed with members of other species.
- Organisms in the same species share a common ancestor.

How do new species arise? New species arise when one existing species splits into two reproductively-isolated populations that go their separate ways. This most commonly happens when the two populations become physically separated from each other (allopatric speciation):

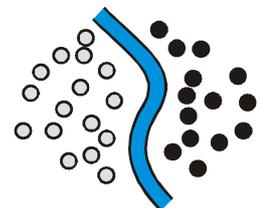
1. Start with an interbreeding population of one species.



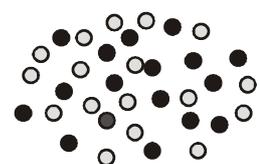
2. The population becomes divided by a physical barrier such as water, mountains, desert, or just a large distance. This can happen when some of the population migrates or is dispersed, or when the geography changes catastrophically (e.g. earthquakes, volcanoes, floods) or gradually (erosion, continental drift). The populations must be reproductively isolated, so that there is no gene flow between the groups.



3. If the environments (abiotic or biotic) are different in the two places (and they almost certainly will be), then different characteristics will be selected by natural selection and the two populations will evolve differently. Even if the environments are similar, the populations may still change by random genetic drift, especially if the population is small. The allele frequencies in the two populations will become different.



4. Much later, if the barrier is now removed and the two populations meet again, they are now so different that they can no longer interbreed. They therefore remain reproductively isolated and are two distinct species. They may both be different from the original species, if it still exists elsewhere.



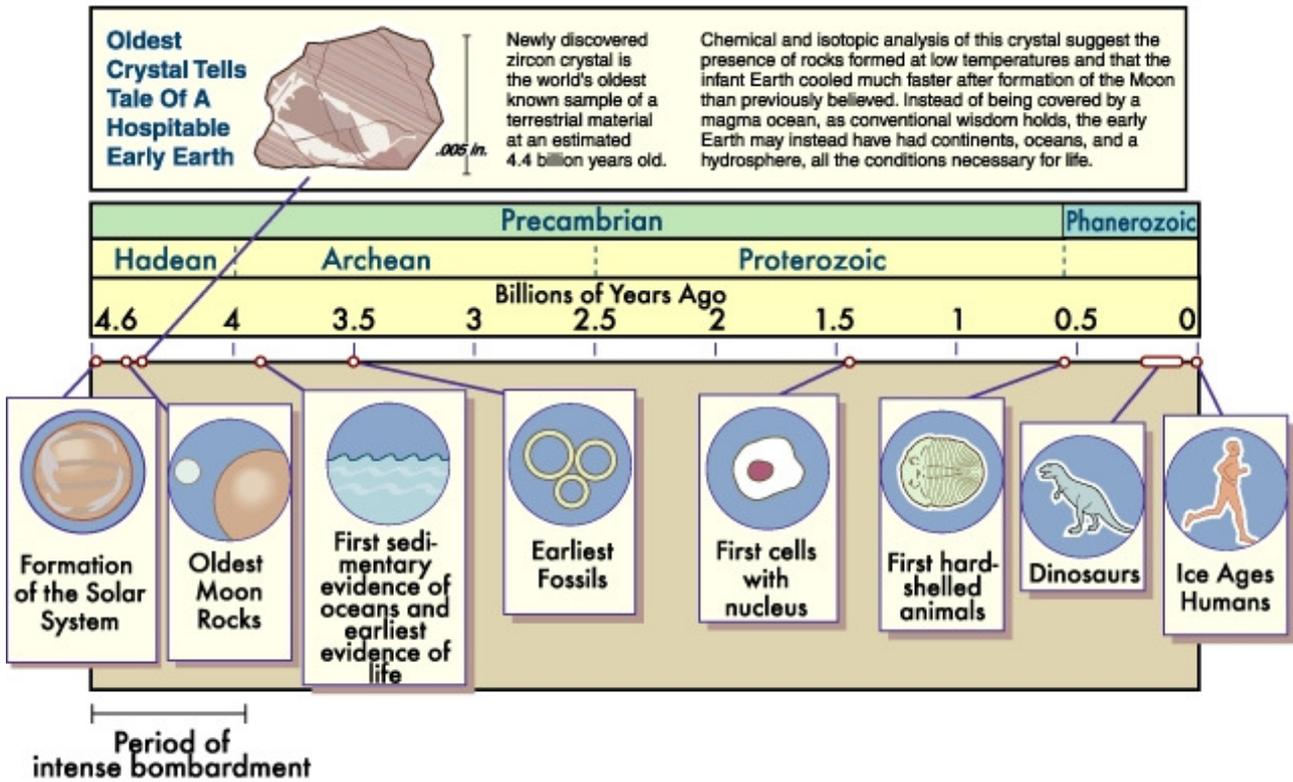
Summary of Speciation

1. A population becomes separated into two groups that are reproductively isolated, so that there is no gene flow between the groups.
2. The two groups' environments are different, so natural selection favours different characteristics.
3. The allele frequencies in the two groups will change in different ways.
4. Eventually the two populations will be unable to interbreed, so will be different species.

It is meaningless to say that one species is absolutely better than another species, only that it is better adapted to that particular environment. A species may be well-adapted to its environment, but if the environment changes, then the species must evolve or die. In either case the original species will become extinct. Since all environments change eventually, it is the fate of all species to become extinct (including our own).

Deep Time and the Origin of life

It takes time to evolve 100 million species and it is now known that the earth is 4,600 million years old. Life (in the form of prokaryotic cells) arose quite quickly, and has existed for around 4,000 million years. These huge spans of time are almost impossible to comprehend, and are often referred to as deep time. This chart illustrates some of the events in the history of the Earth.



No one knows how life arose in the first place, but the conditions in the early Earth were very different from now, and experiments have shown that biochemicals like amino acids and nucleotides could be synthesised from inorganic molecules under primordial conditions. We saw in unit 1 how lipid bilayers can form spontaneously, so perhaps that's how cellular life arose.

Appendix I – Biological Principles

Some basic biological principles from units 1 and 2 can be examined in unit 4.

Unit 1 Biological principles

- Proteins and polysaccharides are made up of monomers that are linked by condensation.
- Many of the functions of proteins may be explained in terms of molecular structure and shape.
- Enzymes are proteins and their rates of reaction are influenced by a range of factors: temperature, the presence of inhibitors, pH and substrate concentration.
- Substances are exchanged by passive or active transport across exchange surfaces. The structure of plasma membranes enables control of the passage of substances across exchange surfaces.

Unit 2 Biological principles

- A species may be defined in terms of observable similarities and the ability to produce fertile offspring.
- Living organisms vary and this variation is influenced by genetic and environmental factors.
- The biochemical basis and cellular organisation of life is similar for all organisms.
- Genes are sections of DNA that contain coded information as a specific sequence of bases.
- During mitosis, the parent cell divides to produce genetically identical daughter cells.
- The relationship between size and surface area to volume ratio is of fundamental importance in exchange.

No other content from units 1 and 2 can be tested in unit 4.

Mathematical Requirements

Biology is a quantitative science, and a reasonable mathematical ability is expected in an A-level biology exam. The unit 4 exam can test any of these mathematical topics:

Calculations

- Use standard form; ratios, fractions and percentages.
- Calculate x^n ; $1/x$; \sqrt{x} ; mean; and standard deviation.
- Calculate percent change and rate of change.
- Calculate circumferences and areas of circles; and surface areas and volumes of cuboids and cylinders when provided with appropriate formulae.
- Use units with prefixes (n, μ , m, k, M, G) and use an appropriate number of significant figures.
- Make estimates of the results of calculations without using a calculator.
- Rearrange equations and substitute numerical values into equations using appropriate units.

Handling data

- Understand the terms mean, median and mode and standard deviation.
- Understand the use of logarithms for quantities that range over several orders of magnitude.
- Construct and interpret frequency tables, bar charts and histograms.
- Use a scatter diagram to identify positive and negative correlation between two variables.
- Plot graphs from data (using appropriate institute of biology conventions) and read data from graphs.
- Understand the principles of sampling as applied to biological data.
- Write a null hypothesis and interpret p -values as the probability of the observed results happening by chance.

Appendix 2 – The Unit 4 Exam

The three A2 biology units are assessed as shown in this table:

Unit	Assessment	Details	Raw marks	UMS marks
Unit 4	1h 30min exam	6-9 short answer questions plus 2 longer questions : 1 HSW and 1 continuous prose.	75	100
Unit 5	2h 15min exam	8-10 short answer questions plus 2 longer questions : 1 data handling (25mk) and 1 synoptic essay (25mk).	100	140
Unit 6	A2 EMPA	2 practical sessions with short written task sheets plus a 1h 15min exam.	50	60
Total				300

Biology is not just about learning facts (though there is a lot to learn): it's largely about understanding principles and being able to apply these principles to unfamiliar situations (which is what happens in real life). It's also important to understand How Science Works, and the role of evaluation and critical thinking. So the A2 biology exams test all these aspects. Of the 75 raw marks in the unit 4 exam, about 22 will be for biological knowledge; 35 will be for applying that knowledge to unfamiliar situations and analysing data; and 18 will be for How Science Works, including planning, analysing and evaluating experiments. So expect lots of questions about data analysis. These are designed to test your knowledge of unit 4 biology in unfamiliar contexts.

How Science Works

You need to understand all the How Science Works words on the next page.

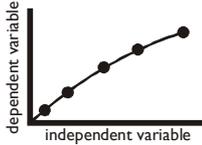
Types of Variable

Dependent Variable

The variable you *measure*, to see how it is affected by the independent variable.

Independent Variable

The variable you *choose to change*, to see how it affects the dependent variable. You may also measure it when you change it.



Confounding Variables

Any variables that could also affect the dependent variable. Confounding variables should be controlled in a fair test.

Control variables

Confounding variables that are kept constant (controlled) during the experiment. If you can't control a variable (such as weather in a field investigation), you should at least monitor it.

Experimental Design

Controlled Experiment (Fair Test)

When all relevant variables are controlled, so that observed changes in the dependent variable must be due to changes in the independent variable.

Control Experiment (Control)

An additional experiment designed to eliminate alternative explanations for the main experiment, and so show that observed changes in the dependent variable must be due only to changes in the independent variable.

Control Group

A group or sample treated in the same way as the experimental group, except for the factor being investigated e.g. a placebo group in a drugs trial. By comparing the results for two groups it can be shown that observed changes in the dependent variable must be due only to changes in the independent variable.

Placebo

A dummy pill, injection or treatment that has no physiological effect (e.g. a sugar pill or saline injection). Used in a clinical trial to allow for the **placebo effect** - the observation that symptoms can improve when patients believe they are being helped.

RCT

The best experimental design for a drug trial. RCT stands for Randomised Controlled Trial, or in more detail, a Randomised, Placebo-Controlled, Double-blind Trial. This design ensures that the trial is **valid** free from **bias**.

- **Randomised** means the study and control groups are allocated randomly
- **Placebo-controlled** means the study group (taking the drug to be tested) is compared to a placebo group (who are given a placebo).
- **Double-blind** means that neither the subjects nor the investigators know who is in the study or placebo groups. This avoids bias.

Protocol

A method or technique that has been shown to produce valid and reliable results.

Hypothesis

A suggested explanation of observations or results that can be tested. Also known as a scientific hypothesis. A good hypothesis can be used to make **predictions**.

Quality of Data

True Value

The real value of a measurement, if it could be measured with no errors at all.

Precise Data

1. Measurements that give similar values when repeated. The replicates therefore have a small **range**.
2. Data measured on sensitive equipment with a suitably fine scale, e.g. 20 mm is more precise than 2 cm.

Reliable Data

Findings that can be repeated. This includes by the original investigator; by other scientists; by other techniques; or those that agree with secondary sources.

Accurate Data

Measurement that are close to the **true value**.

Valid Data

The best quality data, i.e. data that is **precise** and **reliable** and obtained from an **unbiased, controlled** experiment that addresses the stated aim. Valid data is assumed to be accurate.

Evidence

Any data or observations that are used to support a particular hypothesis.

Anecdote

An observation or story from real life. Anecdotes are not evidence and cannot be used to support a hypothesis, but they can be useful to suggest a new testable hypothesis.

Types of Data

Data

(measurements, singular datum)

Quantitative or Numeric Data (numbers)

Continuous Data

can have any value
e.g. 7.34, -294.6, 2×10^5

Discrete Data

only whole numbers
e.g. no. of atoms

Qualitative or Categorical Data (words)

Ordered Data

can be ranked
e.g. small, medium, large

Nominal Data

can't be ranked
e.g. male, female

Errors

Random Errors

Inaccuracies due to mistakes, poor technique, or random variation. Random errors are very common, but can be improved by taking many replicates. Data with a small random error is said to be **precise**.

Systematic Errors

Inaccurate measurements in one direction only, due to poor **calibration** or poor technique. Systematic errors can **not** be improved by taking more replicates. Data with a small systematic error is said to be **reliable**.

Zero Error

A particular kind of systematic error, where the instrument does not return to zero.

Bias

When the observer chooses some results and ignores others, to support a particular view. Also called **cherry picking**.

Anomaly or Outlier

A measurement that falls far outside the expected range and is therefore probably due to experimental error. Anomalies should be rejected, since they skew the mean, but it is very difficult to distinguish between anomalies and normal biological variation.

Calibration

Ensuring that a measuring instrument gives correct readings by fixing known points then constructing a scale between them.

Simple Analysis

Replicates

Repeats of a measurement.

Raw Data

The original measurements or recordings before any manipulation or processing.

Mean or Average

The mid-point of the replicates.
= sum of replicates / N

Range

The highest and lowest replicates, or the interval between them.

Standard Deviation (SD)

A measure of the dispersal of the replicates about the mean. In a normal distribution 68% of the replicates will be within 1 standard deviation of the mean, and 95% will be within 2 standard deviations of the mean.

Standard Error of the mean (SEM)*

A measure of the uncertainty, or error, of a calculated mean. The smaller the standard error, the more reliable the mean.

95% Confidence Interval (CI)*

Another measure of the error of the mean. We can be 95% confident that the true mean lies in the range (mean \pm CI). The top and bottom of this range are called the **confidence limits**.

Statistical Analysis

Correlation (or Association)

When one variable changes with another variable, so there is a relation between them. The strength of a correlation can be measured using a correlation coefficient. A correlation need not be a **causal relation**.

Causal Relation

When changes in one variable cause the changes in another variable. Can only be shown by a controlled experiment.

Statistical Test*

Something that tests whether observed differences or correlations are significant, or just due to chance.

Null Hypothesis*

The statement that is tested by a statistical test. The null hypothesis is fixed for each test, but always says that there is no difference or no association. The null hypothesis has nothing to do with a scientific hypothesis.

P-value*

The result of a stats test, expressed as a probability. It is the probability that the results are due to chance. If $P < 0.05$ then we reject the null hypothesis, otherwise we accept it.