

Photosynthesis

Revision notes · The light-dependent and light-independent reactions, chloroplast structure, pigments and spectra, and the core practical on rate of photosynthesis.

For Pearson Edexcel International AS Biology XBI11 and International A Level Biology YBI11. **A Level only** is IA2 content (Units 4 to 6), not needed for the International AS.

Photosynthesis **Spec 5.1-5.8** **A Level only**

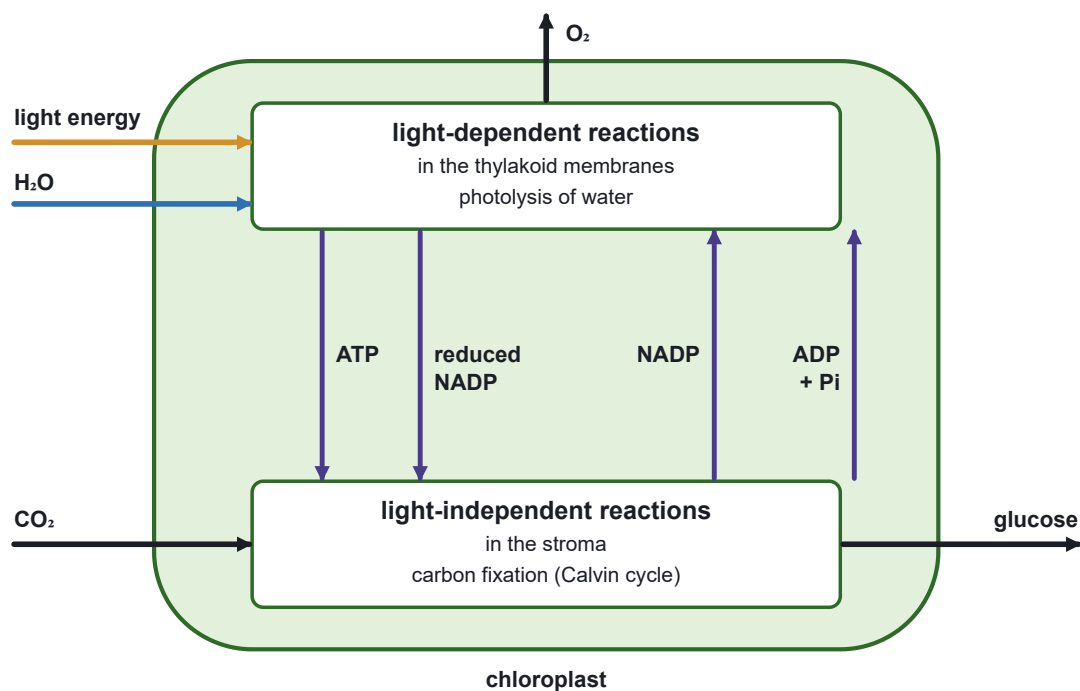
The overall reaction and the role of ATP **Spec 5.1, 5.2**

The overall reaction is a **redox reaction**. Light energy is used to break the strong bonds in water molecules (photolysis). The hydrogen from the water is stored in a fuel, **glucose**, by combining it with carbon dioxide. Oxygen is released into the atmosphere as a by-product.



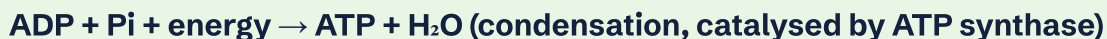
Carbon dioxide is reduced (it gains hydrogen) and water is oxidised (it loses hydrogen and electrons). The oxygen released comes from water, not from carbon dioxide. This can be shown by supplying plants with water labelled with the isotope oxygen-18: the oxygen gas given off is labelled, but the glucose is not. The summary equation above hides this, because 6 water molecules hold only 6 of the 12 oxygen atoms in 6O_2 . A more accurate equation is $6\text{CO}_2 + 12\text{H}_2\text{O} \rightarrow \text{C}_6\text{H}_{12}\text{O}_6 + 6\text{O}_2 + 6\text{H}_2\text{O}$, in which all the oxygen gas comes from water.

The reaction is not carried out in one step. Light energy is first used in the **light-dependent reactions** to make ATP and reduced NADP. These are then used in the **light-independent reactions** to reduce carbon dioxide to sugars.



The **light-dependent reactions** make ATP and **reduced NADP** (a hydrogen carrier) and release oxygen from water; the **light-independent reactions** use them to fix carbon dioxide into glucose.

ATP (adenosine triphosphate) is the immediate source of energy for biological processes. When the terminal phosphate bond is hydrolysed, ATP becomes ADP and inorganic phosphate (Pi) and a small, usable amount of energy is released. This is done in one step, catalysed by ATP hydrolase (ATPase), so energy is available quickly and in amounts the cell can use.



In photosynthesis, the ADP is converted to ATP by **photophosphorylation**: the addition of phosphate to ADP using light energy. This requires energy because the reaction $\text{ADP} + \text{Pi} \rightarrow \text{ATP}$ is endergonic. The ATP made in the light-dependent reactions is hydrolysed in the light-independent reactions to power the reduction of carbon dioxide.

Exam tip: Say that photophosphorylation uses light energy to add phosphate to ADP. Do not say that ATP stores or 'makes' energy: it releases energy when it is hydrolysed.

The light-dependent reactions Spec 5.3

The light-dependent reactions take place on the **thylakoid membranes** of the chloroplast. Chlorophyll and other pigments are arranged in two groups called photosystems: photosystem I (PSI) and photosystem II (PSII). Each photosystem traps light energy and passes it to a reaction centre containing chlorophyll a.

How light energy is trapped

When a photon of light hits chlorophyll, an electron in the chlorophyll is raised to a higher energy level. The electron is said to be **excited**. It leaves the chlorophyll molecule and is passed to an electron carrier. The chlorophyll has been oxidised (it has lost an electron).

Non-cyclic photophosphorylation

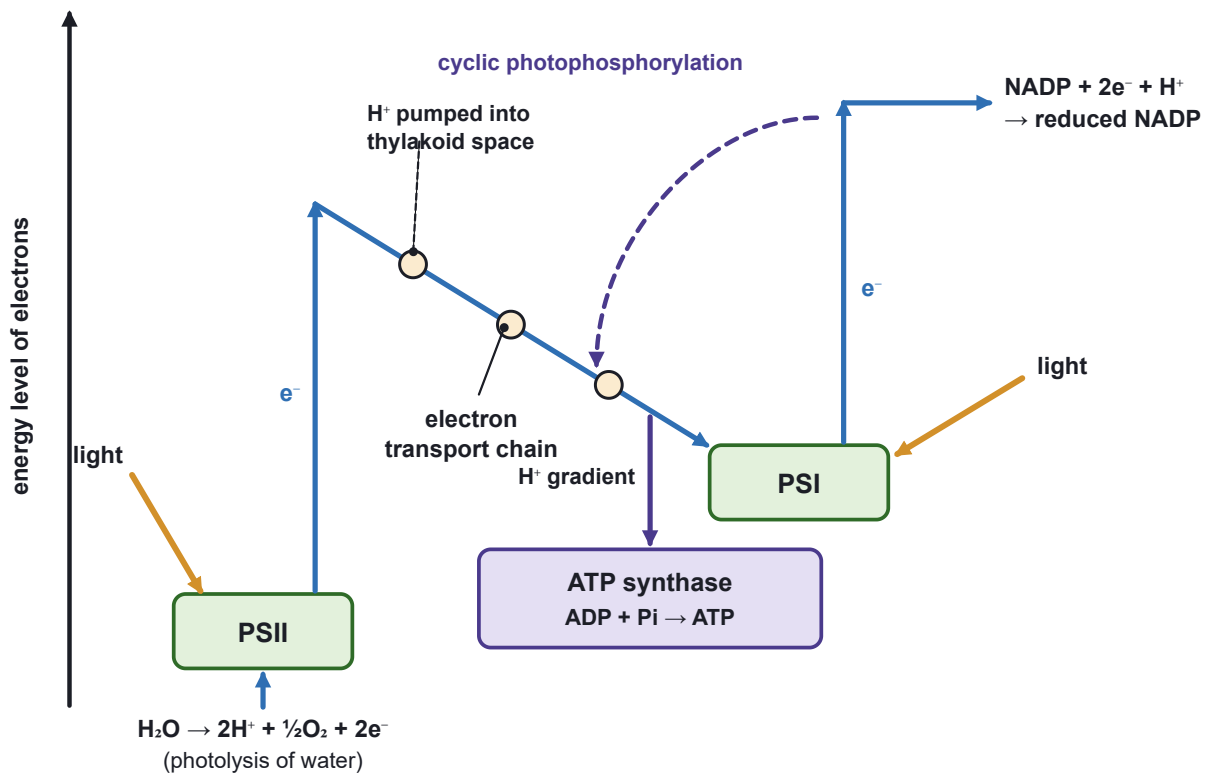
1. Light energy excites two electrons in chlorophyll in photosystem II. They leave the chlorophyll and are taken up by the first electron carrier in the electron transport chain.
2. The electrons pass down the electron transport chain from carrier to carrier, losing energy at each step in a series of redox reactions.
3. The energy lost is used to pump protons (H^+) from the stroma into the thylakoid space. This builds up a proton gradient across the thylakoid membrane.
4. Protons flow back down their concentration gradient into the stroma through ATP synthase. This is **chemiosmosis**. The flow drives the synthesis of ATP from ADP and P_i (photophosphorylation).
5. Light also excites electrons in photosystem I. These electrons are passed to NADP, which also accepts a proton (H^+) from the stroma, and is reduced to **reduced NADP**.
6. The electrons lost from photosystem II are replaced by electrons from water. The enzyme-catalysed **photolysis** of water splits it: $2H_2O \rightarrow 4H^+ + 4e^- + O_2$. The electrons replace those lost from chlorophyll. The H^+ ions contribute to the proton gradient and are used to reduce NADP. The oxygen is a waste product, and diffuses out of the leaf or is used in respiration.

Because electrons flow in one direction, from water through the two photosystems to NADP, this is called non-cyclic photophosphorylation. Its products are ATP, reduced NADP and oxygen.

Cyclic photophosphorylation

In cyclic photophosphorylation only photosystem I is used. Excited electrons leave chlorophyll, pass along an electron transport chain and return to the same chlorophyll molecule. Energy lost along the chain is used to make ATP by chemiosmosis. No NADP is reduced, no water is split and no oxygen is made. The electrons are recycled, so no replacement electrons are needed.

Cyclic and non-cyclic photophosphorylation		
Feature	Non-cyclic	Cyclic
Photosystems used	Photosystem II and I	Photosystem I only
Source of electrons	Water (photolysis)	The same chlorophyll (electrons return)
ATP made	Yes	Yes
Reduced NADP made	Yes	No
Oxygen made	Yes	No



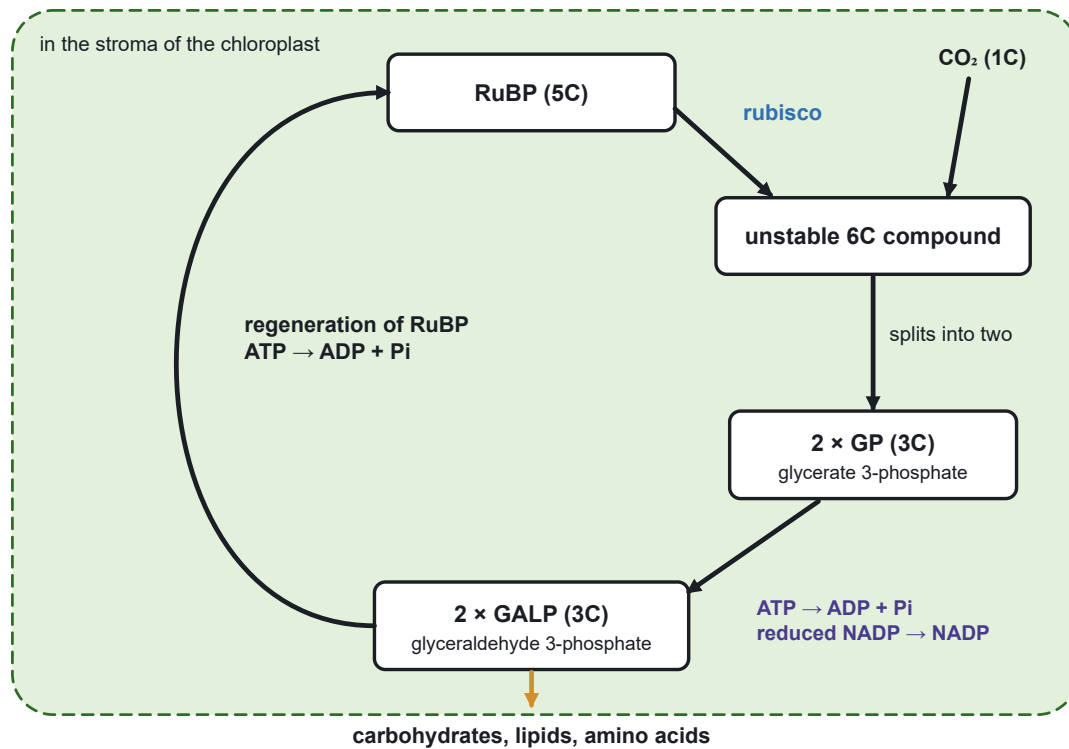
The **Z-scheme** of the light-dependent reactions: electrons are raised to a higher energy level twice, with ATP made as they pass down the chain.

Common mistake: Photolysis is not the splitting of carbon dioxide. Oxygen released in photosynthesis comes from water.

The light-independent reactions (Calvin cycle) Spec 5.4

The light-independent reactions are a cycle of enzyme-controlled reactions called the **Calvin cycle**. They take place in the **stroma** of the chloroplast. They do not need light directly, but they depend on the ATP and reduced NADP made in the light-dependent reactions, so they stop soon after the light is switched off.

1. **Carbon fixation**: carbon dioxide diffuses into the stroma and combines with the five-carbon compound ribulose biphosphate (**RuBP**). The reaction is catalysed by the enzyme ribulose biphosphate carboxylase-oxygenase (**rubisco**). The unstable six-carbon compound formed splits immediately into two molecules of the three-carbon compound glycerate 3-phosphate (**GP**).
2. **Reduction**: GP is reduced to glyceraldehyde 3-phosphate (**GALP**, a triose phosphate) using hydrogen from reduced NADP and energy from ATP. Reduced NADP is oxidised back to NADP, and ATP to ADP and Pi, which return to the light-dependent reactions.
3. **Regeneration**: most of the GALP (five of every six molecules) is used, with energy from ATP, to regenerate RuBP so that the cycle can continue.
4. **Synthesis**: the remaining GALP is used to make glucose and other organic molecules. Two GALP molecules are needed to make one hexose sugar, so the cycle must turn six times to fix the six carbon atoms needed for one glucose molecule.



The **Calvin cycle**: carbon fixation by rubisco, reduction of GP to GALP, and regeneration of RuBP.

Uses of the products (5.4(ii))

The products of photosynthesis are **simple sugars**. These are used by plants, animals and other organisms in respiration, to release energy, and in the synthesis of new biological molecules.

Biological molecules made from the products of photosynthesis	
Molecule made	How the products of the Calvin cycle are used
Polysaccharides	Glucose is made from GALP, then joined by condensation to make starch (storage) and cellulose (cell walls)
Lipids	Glycerol is made from GALP, and fatty acids from GP (via acetyl CoA), then combined to form triglycerides
Amino acids and proteins	GP provides the carbon skeletons. Nitrogen comes from nitrate ions absorbed from the soil. Amino acids are joined by condensation to form proteins
Nucleic acids	Sugars provide the pentose sugars (ribose, deoxyribose) and carbon for bases. Phosphate groups come from phosphate ions in the soil

Animals and other organisms (such as fungi and many microorganisms) obtain these sugars and the molecules made from them by feeding, and use them in the same ways.

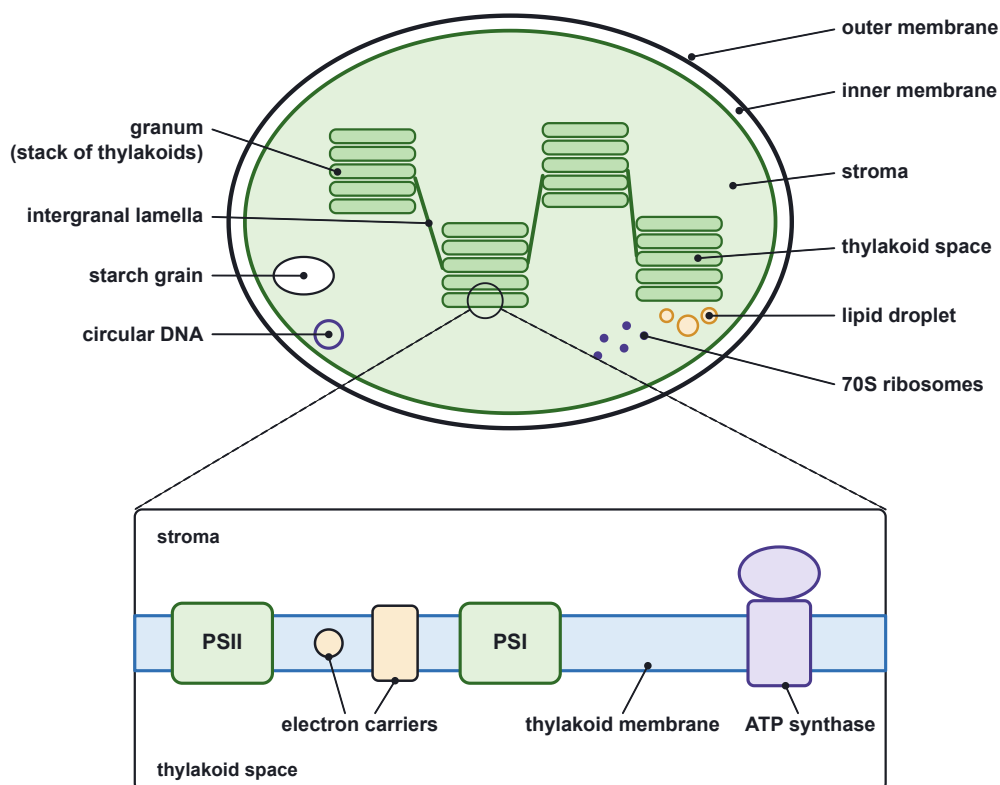
Exam tip: If asked what happens to GP and RuBP levels when conditions change, work through the cycle. In darkness, ATP and reduced NADP run out, so GP cannot be reduced; GP rises and RuBP falls. With less carbon dioxide, RuBP cannot be used; RuBP rises and GP falls.

Chloroplast structure and function Spec 5.5

Chloroplasts are found mainly in palisade mesophyll cells. They are biconvex discs about 2 to 10 μm long. Each is surrounded by a **chloroplast envelope** of two phospholipid membranes, which controls the entry and exit of substances such as carbon dioxide, ATP and sugars.

Chloroplast structure related to function

Structure	Description	Role in photosynthesis
Thylakoid membranes	Flattened fluid-filled sacs, with a thylakoid space inside	Contain chlorophyll and other pigments in photosystems, electron carriers and ATP synthase. Site of the light-dependent reactions. The small space inside allows a proton gradient to build up quickly
Grana (singular granum)	Stacks of thylakoids	Give a very large surface area for pigments, so more light is absorbed. Stacks are arranged to capture light efficiently
Intergranal lamellae	Thylakoid membranes joining one granum to the next	Link grana, and keep them in position
Stroma	Colourless fluid-filled matrix surrounding the grana	Contains the enzymes (including rubisco) for the light-independent reactions, starch grains, lipid droplets, and the chloroplast's own DNA and ribosomes
Starch grains	Insoluble stores in the stroma	Temporary store of the sugars made in the Calvin cycle
Chloroplast DNA and ribosomes	A small circular DNA molecule and 70S ribosomes	Allow chloroplasts to make some of their own proteins, for example some rubisco subunits



A chloroplast, with a magnified view of the **thylakoid membrane** where the light-dependent reactions happen.

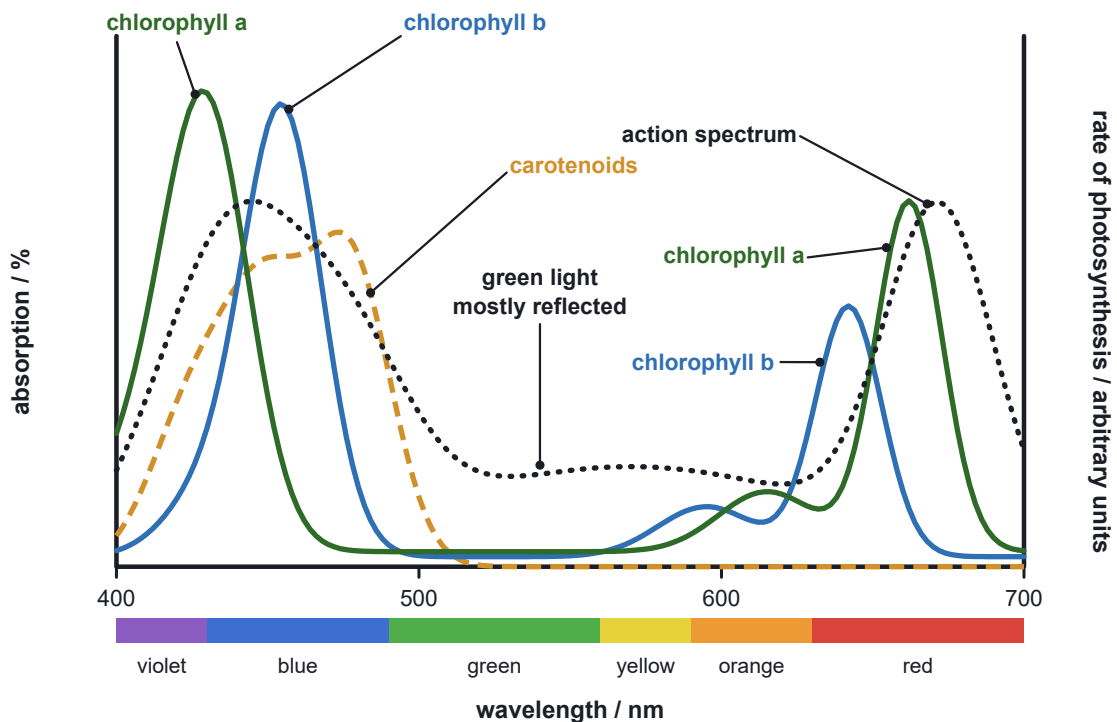
The two stages of photosynthesis are in different places but close together. The ATP and reduced NADP made on the thylakoid membranes are used straight away in the stroma next door, so little energy is lost in transport.

Pigments, spectra and chromatography Spec 5.6, 5.7

Absorption spectrum	A graph showing how much light of different wavelengths is absorbed by a pigment (or by a leaf extract).
Action spectrum	A graph showing the rate of photosynthesis (for example oxygen released or carbon dioxide taken up) at different wavelengths of light.

Chloroplasts contain a mixture of photosynthetic pigments. **Chlorophyll a** and **chlorophyll b** absorb mostly red and blue-violet light, and reflect green light. Carotenoids (for example carotene and xanthophyll) absorb mostly blue-violet and blue-green light. Different pigments absorbing different wavelengths mean that more of the available light is used.

The action spectrum has a similar shape to the combined absorption spectra of the pigments. This shows that the wavelengths that are absorbed are the ones used in photosynthesis. The action spectrum is not identical to the absorption spectrum, because some light absorbed by accessory pigments is passed on to chlorophyll a with some loss of efficiency, and not all absorbed light is used in photosynthesis.



Absorption spectra of the pigments and the **action spectrum** share the same peaks in blue-violet and red light.

Common mistake: Do not say that an action spectrum shows how much light is absorbed. It shows the rate of photosynthesis.

Separating pigments by chromatography (5.7)

1. Grind fresh leaves with a little propanone (or ethanol) and sand to extract the pigments.
2. Draw a pencil line (the origin) about 2 cm from the bottom of a chromatography paper strip (or a thin-layer chromatography plate). Use pencil because ink would also dissolve in the solvent.
3. Apply a small spot of extract on the line and let it dry. Repeat several times to build up a concentrated spot.
4. Stand the paper in a container with a shallow layer of solvent (a mixture such as petroleum ether and propanone) below the origin line. Cover the container.

5. When the solvent front is near the top, remove the paper and mark the solvent front at once with a pencil before it evaporates.
6. Measure the distance from the origin to the centre of each pigment spot and from the origin to the solvent front.

The pigments separate because they have different solubilities in the solvent and different affinities for the paper (or plate). A pigment that is more soluble in the solvent, and sticks less strongly to the paper, travels further.

$$R_f = \text{distance moved by the pigment} \div \text{distance moved by the solvent front}$$

The R_f value has no units and is always less than 1. The same pigment gives the same R_f value in the same solvent and under the same conditions, so R_f values can be compared with published values to identify pigments. Carotene travels furthest (highest R_f), followed by xanthophyll, then chlorophyll a, then chlorophyll b (lowest R_f of the four).

Calculating an R_f value

A pigment spot is 5.4 cm from the origin. The solvent front is 7.2 cm from the origin. Calculate the R_f value of the pigment.

1. $R_f = \text{distance moved by the pigment} \div \text{distance moved by the solvent front}$
2. $R_f = 5.4 \div 7.2$

Answer: $R_f = 0.75$ (no units)

Maths skill: Measure to the centre of each spot, and measure both distances from the same origin line. R_f has no unit, and a value above 1 means a measuring slip.

Exam tip: If asked about risks or good practice in chromatography: handle propanone and other solvents in a well-ventilated area away from flames, wear eye protection, and keep the lid on the container so the atmosphere in the tank stays saturated with solvent vapour.

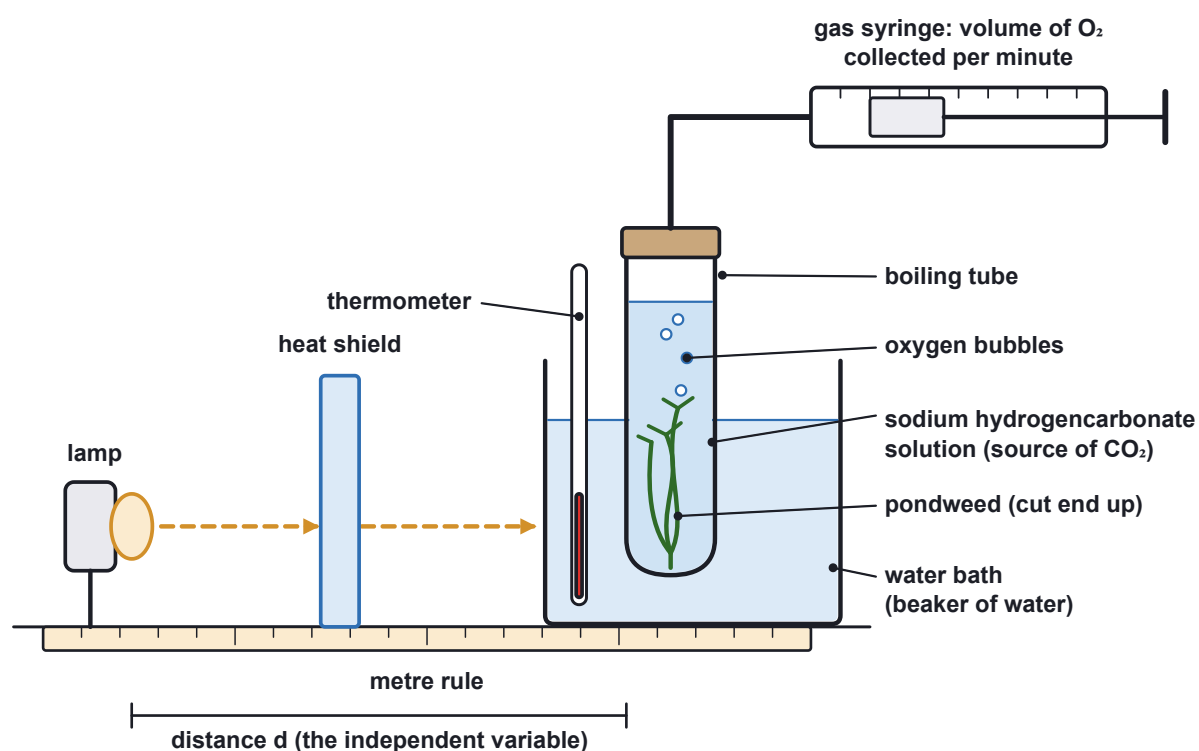
Core practical 10: factors affecting the rate of photosynthesis Spec 5.8

Core practical: Core practical 10: investigate the effects of light intensity, light wavelength, temperature and availability of carbon dioxide on the rate of photosynthesis using a suitable aquatic plant.

A piece of aquatic plant (for example *Cabomba* or *Elodea*) is cut at the stem under water and placed in a boiling tube or beaker with the cut end upwards. Oxygen released in photosynthesis forms bubbles at the cut end. The rate of photosynthesis is estimated from the number of bubbles per minute, or more accurately from the volume of gas collected in a capillary tube or gas syringe in a set time.

Variables in the investigation

Factor (independent variable)	How it is varied	What must be controlled
Light intensity	Move a lamp to different measured distances from the plant (intensity is proportional to $1 \div \text{distance}^2$)	Temperature (place a heat shield, such as a beaker of water, between lamp and plant), carbon dioxide concentration, plant mass, wavelength
Light wavelength	Use coloured filters, or LEDs of different colours, at a fixed distance	Light intensity (check it with a light meter), temperature, carbon dioxide concentration
Temperature	Use a water bath at several set temperatures (for example 10 to 40 °C), checked with a thermometer	Light intensity, carbon dioxide concentration, plant mass; allow the plant to equilibrate before counting
Carbon dioxide availability	Use sodium hydrogencarbonate solutions of different concentrations (for example 0.00 to 0.10 mol dm ⁻³), which release carbon dioxide	Light intensity, temperature, plant mass, volume of solution



Measuring the rate of photosynthesis as the volume of oxygen collected per minute at different lamp distances d (the water bath and heat shield keep the temperature constant).

Method and good practice

- Leave the plant under the lamp for a few minutes to adjust to each new condition (equilibrate), then count bubbles or measure gas volume for a fixed time such as 2 or 5 minutes.
- Repeat at least three times at each value of the independent variable and calculate a mean, ignoring anomalous results.
- Use the same plant (or pieces cut to the same length and mass) throughout, and the same volume of solution in the tube.

- A control with no plant, or with a plant in the dark, shows that bubbles come from the plant's photosynthesis.

Processing results

Rate = volume of oxygen collected $\div$ time, in $\text{cm}^3 \text{ min}^{-1}$. Plot rate against the independent variable. Light intensity can be plotted against $1 \div \text{distance}^2$, because intensity is inversely proportional to the square of the distance from a point source. A graph that levels off shows that another factor has become limiting.

Calculating a rate and a mean

In three trials at 20 cm from the lamp, a plant gave off 0.9, 1.0 and 1.1 cm^3 of oxygen in 5 minutes. Calculate the mean rate of photosynthesis in $\text{cm}^3 \text{ min}^{-1}$.

1. Mean volume = $(0.9 + 1.0 + 1.1) \div 3 = 1.0 \text{ cm}^3$
2. Rate = volume $\div$ time = $1.0 \div 5$

Answer: $0.20 \text{ cm}^3 \text{ min}^{-1}$

Maths skill: Quote the unit of rate ($\text{cm}^3 \text{ min}^{-1}$), and say how many readings the mean was calculated from.

Risks: the lamp and mains electricity are used near water, so keep the socket and lamp away from spills. The lamp can get hot; allow it to cool before moving it. Wash hands after handling plants and pond water.

Exam tip: Light, temperature and carbon dioxide can each be limiting. In an exam, say which factor is limiting at the point where a graph levels off, and why the other factors are not.

Quick check

1. Where does the oxygen released in photosynthesis come from?
2. Name the enzyme that catalyses the fixation of carbon dioxide by RuBP.
3. Which stage of photosynthesis makes reduced NADP?
4. What is the formula for the R_f value?

Quick check answers

Photosynthesis

1. From the photolysis of water.
2. Rubisco (ribulose biphosphate carboxylase-oxygenase).
3. The light-dependent reactions (non-cyclic photophosphorylation).
4. Distance moved by the pigment $\div$ distance moved by the solvent front.