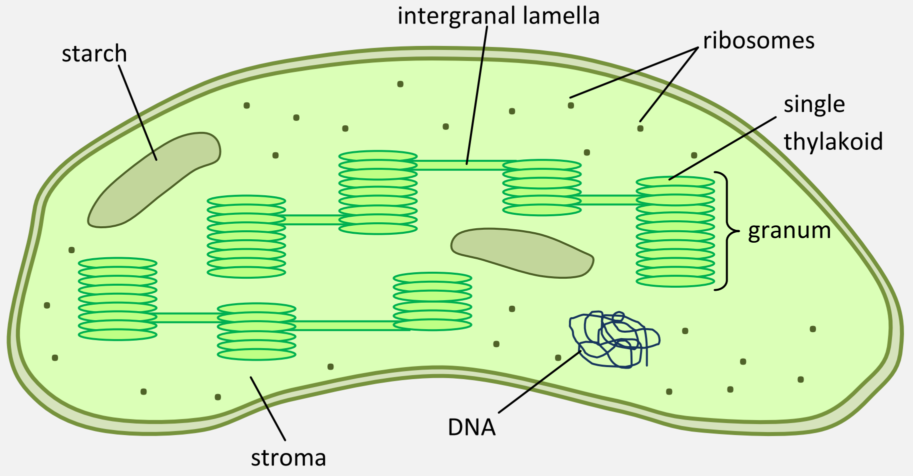
F214 Communication, homeostasis and energy

Unit 3: Photosynthesis

Chloroplasts

The process of **photosynthesis** is one which converts light energy into chemical energy to synthesis large organic molecules from smaller, inorganic substrates. Plants and other photosynthetic organisms are therefore **autotrophic** (they make their own food). There are two main stages of photosynthesis, which both occur inside the **chloroplast**



Inside chloroplasts are stacks of flattened membrane compartments called **grana** (each single compartment called a **thylakoid**). Thin membranal extensions connect one granum to another, these are called **intergranal lamellae**. The fluid surrounding the grana is called the **stroma**. The chloroplast contains its own DNA and many ribosomes, so that is can make proteins

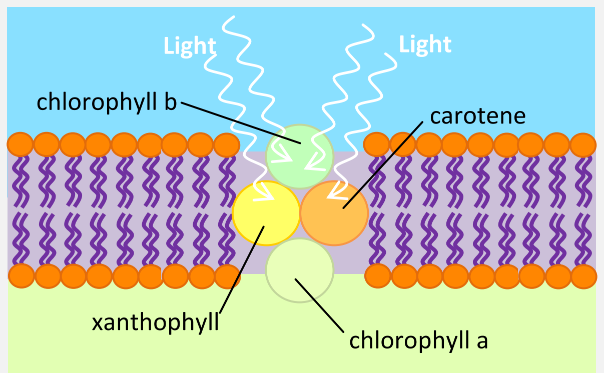
Adaptations of the organelle for its function during photosynthesis include:

- the granal membranes provide a large surface area for the attachment of the photosynthetic pigments (chlorophylls and carotenoids), electron carriers and enzymes for the light-dependent reactions
- a network of proteins in the grana hold the pigments in a very precise manner that forms the photosystems allowing for maximum absorption of light
- the membranes have ATP synthase attached to them which via chemiosmosis help to manufacture ATP
- the fluid of the stroma holds all of the enzymes needed to carry out the light-independent reactions

Photosynthetic Pigments and Photosystems

Embedded in the thylakoid membranes are coloured compounds called **photosynthetic pigments** which absorb light of certain wavelengths and reflect other light. Pigments are found in large numbers arranged into **photosystems**

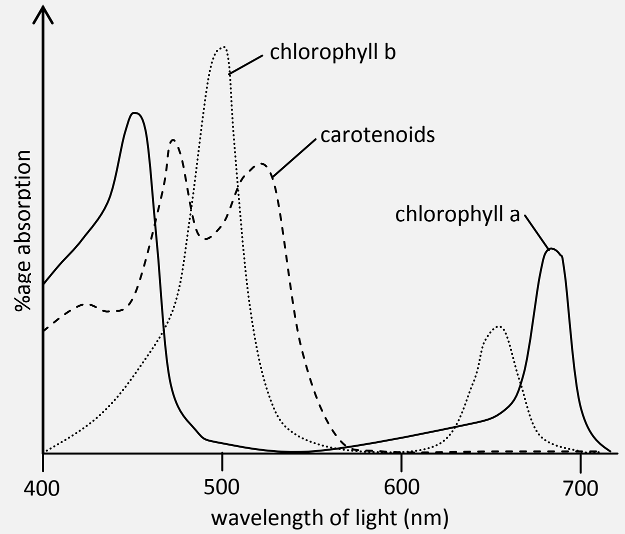
There are two types of pigment found in photosystems. The chlorophylls are **chlorophyll b** and **chlorophyll a**. The carotenoids are **xanthophyll** and **carotene**. Chlorophyll b, xanthophyll and carotene are known as secondary pigments, closest to the stroma side of the membrane, and absorb the light, exciting a pair of electrons to be passed to chlorophyll a the primary pigment

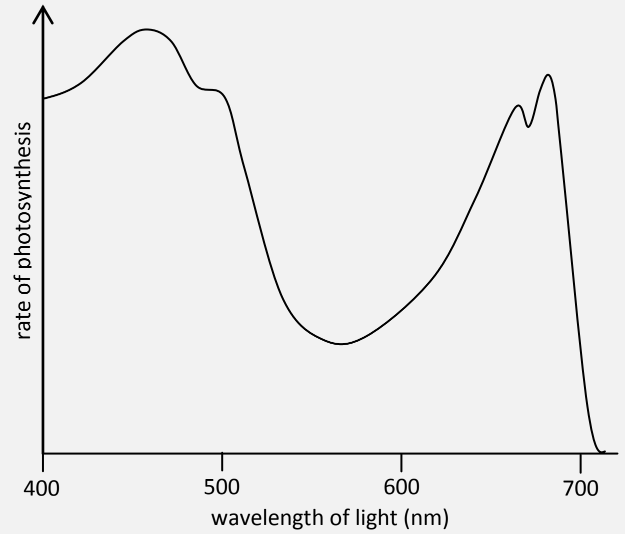


Pigment	Colour	Peak absorption wavelength (nm)	Function in photosynthesis
Chlorophyll a	Yellow-green	430, 662	Absorbs red and blue-purple light
Chlorophyll b	Blue-green	453, 642	
Carotene	Orange	450	Absorb purple light, protect chlorophylls from damage from light and oxygen
Xanthophyll	Yellow	450-470	

Absorption Spectra and Action Spectrum

A calorimeter can be used to measure the light absorption of different photosynthetic pigments. Findings are plotted onto graphs called **absorption spectra** and the effectiveness of pigments is shown on the **action spectrum** which shows rate of photosynthesis at different wavelengths of light. The table at the bottom of the page summarises the absorption spectra by stating the colours/wavelengths each pigment best absorbs

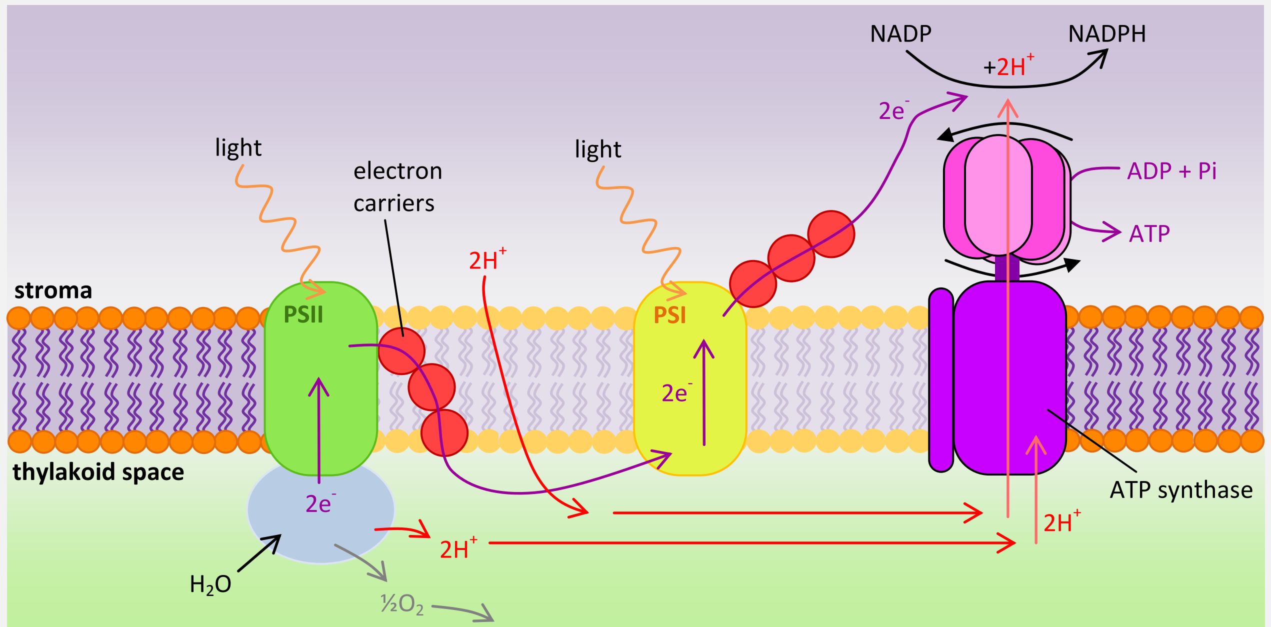




Light-Dependent Reactions

The first of two stages in photosynthesis is the **light-dependent** stage, which takes place in the thylakoid membranes. Two photosystems are used in this stage: **photosystem II** and **photosystem I**, as well as the many **ATP synthase** enzymes embedded in the membranes to manufacture ATP

Firstly, there is **photolysis** of water (splitting using light photons). Two molecules of water – $2H_2O$ – are split into four electrons, four protons and one pair of oxygen atoms. This occurs at photosystem II (which is the first photosystem to be used). The oxygen is actually a waste product and mostly leaves the leaf through the stomata, although some oxygen remains in the plant to be used for aerobic respiration



The electrons however, are accepted by the PSII, and the light energy excites the electrons causing them to move between the pigments until they are accepted by an **electron carrier** which passes the electrons through the membrane, carrier-to-carrier, until one passes the electrons to photosystem I (PSI)

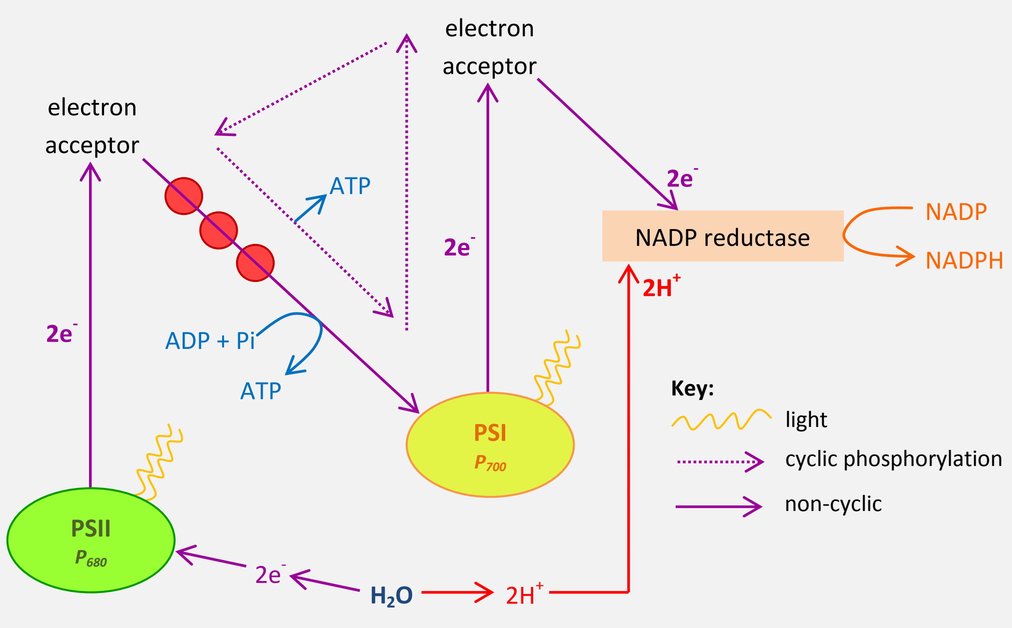
The movement of electrons between electron carriers (**cytochromes**) releases energy, which is used to actively pump protons across the membrane against the gradient

The electrons are accepted by more carriers from PSI and are eventually taken to an enzyme called **NADP reductase** which reduces a coenzyme called **NADP** (nicotinamide adenine dinucleotide phosphate). The hydrogen ions (protons) produced from the photolysis of water remain in the thylakoid space, in order to build up a heavy concentration within the thylakoid. This maintains a constant concentration gradient down from the thylakoid to the stroma (where there is a weaker concentration). This means that when protons are pumped into the thylakoid space between photosystems due to the release of energy from electron carriers (this release of energy and movement of protons is called **chemiosmosis**), protons always flow back over the membrane down the gradient through specialised channel proteins associated with ATP synthase. As protons flow through these channels, ATP synthase is activated and adds one phosphate group to a molecule of ADP to produce ATP (this is **photophosphorylation**)

As hydrogen ions flow back across the membrane into the stroma, they meet up with the coenzyme NADP and the electrons received from the electron carriers. The electrons and protons (two of each) combine to form two hydrogen atoms, and then the enzyme NADP reductase catalyses the reaction between NADP and the hydrogen to produce reduced NADP (**NADPH** or $NADPH_2$). NADPH is one of the products of this stage and is used as the main substrate for the second stage of photosynthesis

Cyclic and Non-Cyclic Photophosphorylation

During the light-dependent reactions, photophosphorylation occurs – this is the addition of an inorganic phosphate group (Pi) to a molecule of ADP to produce ATP in the presence of light. There are two types of photophosphorylation found in this series of processes



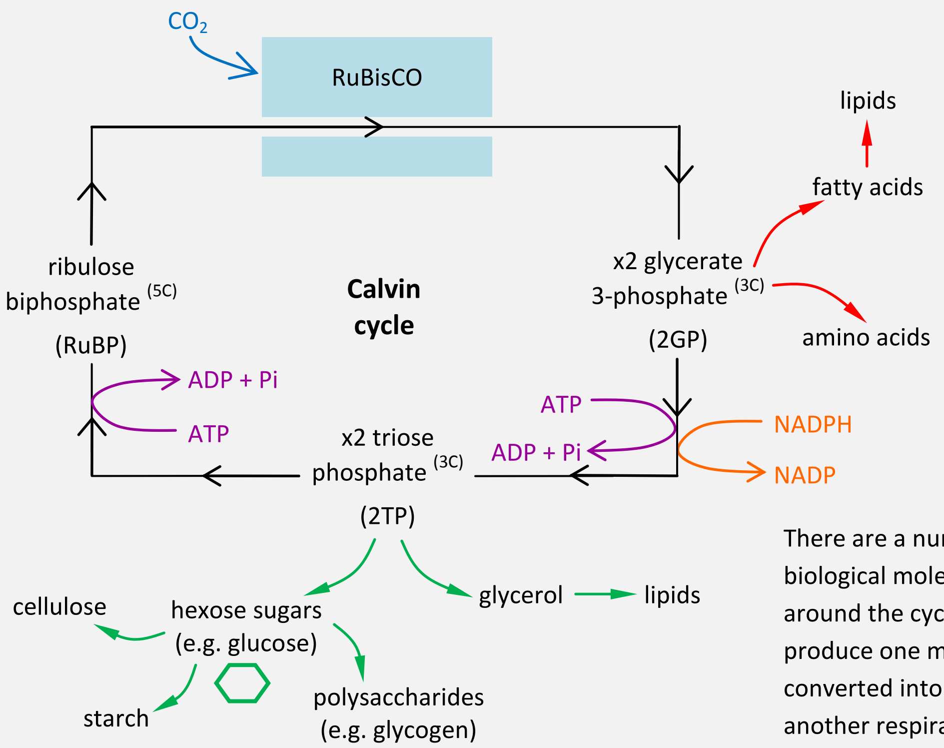
There is **cyclic photophosphorylation** which occurs only using photosystem I. When PSI absorbs a photon of light, chlorophyll a ends up emitting an excited electron which is passed around a chain of carriers before being returned to PSI again. The energy released by the movement of the electrons here is used to generate ATP

Non-cyclic photophosphorylation uses both PSII and PSI. As light excites PSII an electron is passed onto a chain of electron carriers which pass it to PSI. PSII regains its lost electron from the electrons produced by the photolysis of water. The electron then moves from PSI through carriers again and onto NADP reductase. The movement of electrons generates a **proton motive force** which pumps protons through the membrane, and as they diffuse back through channels connected to ATP synthase, ATP is made

Light-Independent Reactions

The second, and final, stage of photosynthesis is known as the **light-independent** stage and does not need light to take place, although without the products of the light-dependent reactions (NADPH and ATP) these reactions cannot occur, so without light this stage also quickly stops working

These reactions take place in the stroma of the chloroplast. The NADPH and ATP produced are already in the stroma, and all else that is needed is carbon dioxide, which enters the leaf by diffusing through the stomata. CO_2 enters the leaf and diffuses through the air spaces in the spongy mesophyll layer, and then through the palisade layer. Then it diffuses through the cellulose cell wall, the cell surface membrane, they cellular cytoplasm and then the chloroplast double membrane into the stroma. The series of reactions here is collectively known as the **Calvin cycle**



1. A molecule of **ribulose bisphosphate** (RuBP), a five-carbon compound, has carbon dioxide added to it by the enzyme **RuBisCO**
2. This forms an unstable six-carbon compound which splits into two molecules of **glycerate phosphate** (GP)
3. The two molecules of GP are phosphorylated (using a phosphate from ATP) and reduced (using hydrogens from NADPH) to form two molecules of **triose phosphate** (TP), also a three-carbon compound
4. Ten of every twelve molecules of TP produced are phosphorylated again and recycled into six molecules of RuBP, and the other two TP go on to be made into other biological molecules (e.g. glucose)

There are a number of opportunities for the Calvin cycle to produce other biological molecules. One sixth of TP produced is not recycled to go back around the cycle, but is modified most often into **glucose** (two TP combine to produce one molecule of glucose) and other hexose sugars. TP can also be converted into glycerol, which combined with fatty acids can form lipids, another respiratory substrate

Also, sometimes molecules of GP are used to form amino acids and fatty acids. Amino acids themselves might be respired to produce energy, and fatty acids become lipids when combined with glycerol from TP. In summary the events of both stages of photosynthesis can be expressed as:

$$6CO_2 + 6H_2O \rightarrow C_6H_{12}O_6 + 6O_2$$

RuBisCO

The enzyme **ribulose bisphosphate carboxylase oxygenase** (RuBisCO) is used in the Calvin cycle to add carbon dioxide to ribulose bisphosphate, but as RuBisCO's name suggests, it is also capable of combining RuBP with oxygen. When this occurs, a process called **photorespiration** takes place. Photorespiration actually undoes a lot of the work that has taken place in photosynthesis so far, so severely lowers photosynthesis efficiency. It also leads to the formation of toxic hydrogen peroxide

When RuBP and oxygen come together, two products are formed: **phosphoglycerate** (PGA) which simply gets converted back into RuBP and re-enters the Calvin cycle so is not too problematic; and **phosphoglycolate** (PPG) which takes a long series of reactions to be reusable again. PPG must leave the chloroplast and be converted several times until it reaches a compound which can re-enter the stroma and enter the Calvin cycle once more. So evidently, this function of RuBisCO is not very efficient and slows down photosynthesis significantly when there is too low a concentration of carbon dioxide in relation to normal atmospheric concentration (allowing oxygen to fit into the enzyme's complex more often)

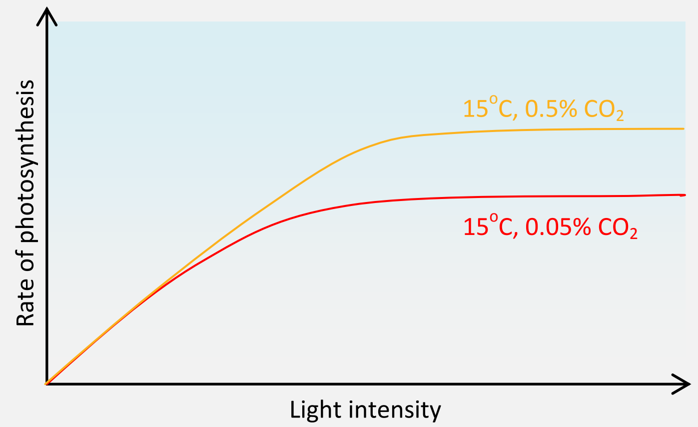
Limiting Factors

A **limiting factor** is any quality of the environment which is the least favourable variable and so impairs the rate of photosynthesis or prevents optimal rate of photosynthesis

Although temperature does not really affect the light-dependent stage, a stable temperature is crucial during the light-independent stage where the enzyme action of RuBisCO is fundamental. Temperatures which fluctuate alter enzyme action, and those which are too high or too low can permanently **denature** enzymes. However, slight increases in temperature do increase enzyme activity. The increase in photosynthesis due to temperature increase depends on the light intensity (when there is more light, increasing temperature has a bigger effect)

Increasing the carbon dioxide concentration does increase the rate of photosynthesis, up until a point where either light intensity or the temperature become the limiting factor

Increasing light intensity generally increases the rate of photosynthesis because more stomata open so more carbon dioxide enters the leaf, more light can be trapped by chlorophyll to excite electrons during the light-dependent reactions, and there is more efficient photolysis of water, all of which will contribute to increasing the rate



Investigating the Rate of Photosynthesis

The rate of photosynthesis is measured using a **photosynthometer** which measures the production of oxygen in a plant. Certain limiting factors (usually light intensity, but also temperature) are adjusted to measure the rate of oxygen production under the different conditions

Bubbles of oxygen collect in a capillary tube one end of the photosynthometer, and when an agreed amount of oxygen has been produced over a controlled time period, a point can be market on a scale where the bubbles reached and the experiment repeated under a different condition

Chromatographic Separation of Photosynthetic Pigments

A **chromatogram** can be used to separate the different photosynthetic pigments in a photosystem. Chromatography is a process whereby a mixture of materials are separated by allowing different particles to 'move' (in what is known as the mobile phase) along a chromatographic strip and fix along a certain place (in what is known as the stationary phase), thus partitioning the different particles