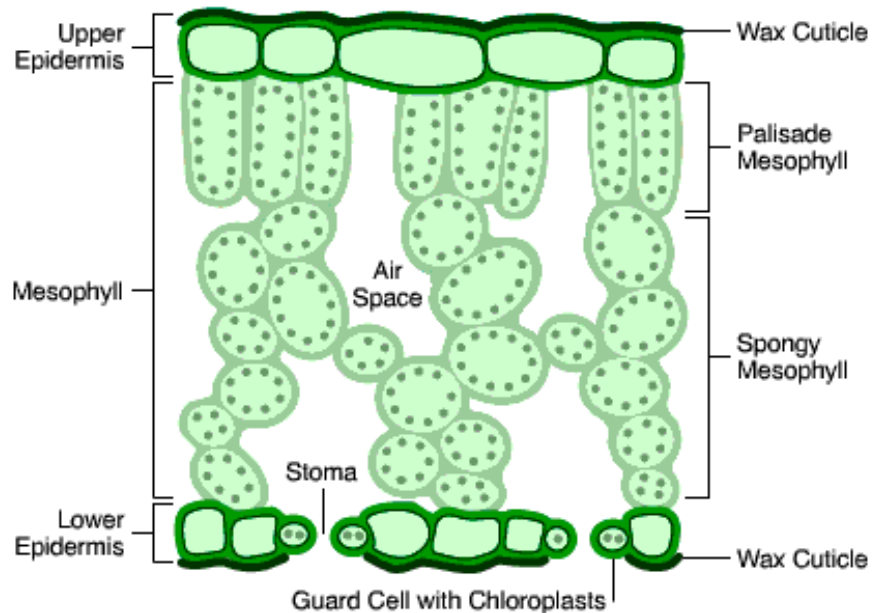


Photosynthesis

Lesson outline

- Introduction
- Structure function of leaf, palisade cell and chloroplast
- Two stages of photosynthesis:
 - Light dependent stage; site, requirements, steps involved, role of pigments, interpret absorption and action spectra of pigments,
 - Light independent stage; site, requirements, steps involved,
- Separating and identifying chloroplast pigments,
- C4 adaptations

Photosynthetic structures: the dicot leaf



- The leaf is the main photosynthetic organ of a plant and it is adapted to facilitate the efficient uptake and absorption of the materials required for photosynthesis
 - **Large surface area** for light absorption
 - Leaves are **arranged to minimise overlapping**
 - **Thin lamina** – short diffusion distance
 - **Transparent cuticle/epidermis** – light can pass through to photosynthetic mesophyll cells
 - The leaf possess **stomata** which open to permit the entry of carbon dioxide into the leaf which then diffuses into the mesophyll cells where photosynthesis occurs
 - The leaf possess veins which contain the **xylem** that brings water from the root and stem. The water moves out of the xylem into neighbouring mesophyll cells
 - The veins also contain **phloem** which carries sucrose and other assimilates away from the leaf
 - Long narrow upper **mesophylls packed with chloroplasts**
 - **Air spaces** in lower mesophyll to allow diffusion of oxygen and carbon dioxide

describe the structure of a dicotyledonous leaf, a palisade cell and a chloroplast and relate their structures to their roles in photosynthesis

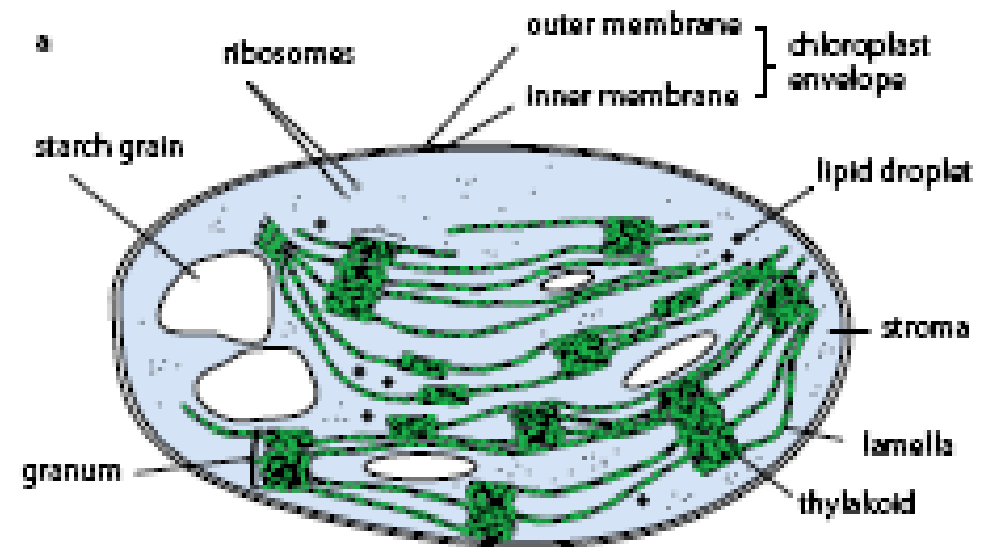
Photosynthetic structures: the palisade cell

- The cells are closely packed together so as to enable them absorb more incident light
- They are arranged near or close to the upper surface of leaf so as to maximize light interception ;
- They are arranged at right angles to leaf surface -- to reduce number of light absorbing walls ;
- They are cylindrical in shape and this allows for air spaces between cells; these air spaces act as reservoir of carbon dioxide ;
- The cells have a large surface area for gas exchange ;
- The cell walls thin and thus it creates short diffusion pathway for gases entering into the cell
- They possess large vacuole which pushes chloroplasts to edge of cell; thus the chloroplasts are on the periphery of the cell and so they can to absorb light more efficiently ;
- They possess a large number of chloroplasts so as to maximise light absorption ;
- The chloroplasts can move within cells towards light and thus maximize light absorption
- The chloroplasts can move away from high light intensity so as to to avoid damage

describe the structure of a dicotyledonous leaf, a palisade cell and a chloroplast and relate their structures to their roles in photosynthesis

Photosynthetic structures: the chloroplast

- Double membrane enclosure
- The interior contains a ground substance called *stroma* which contains enzymes of the Calvin cycle, sugars and organic acids involved in the light independent reactions.
- Stroma also contains 70s ribosomes, DNA loop, lipid droplets and starch grains

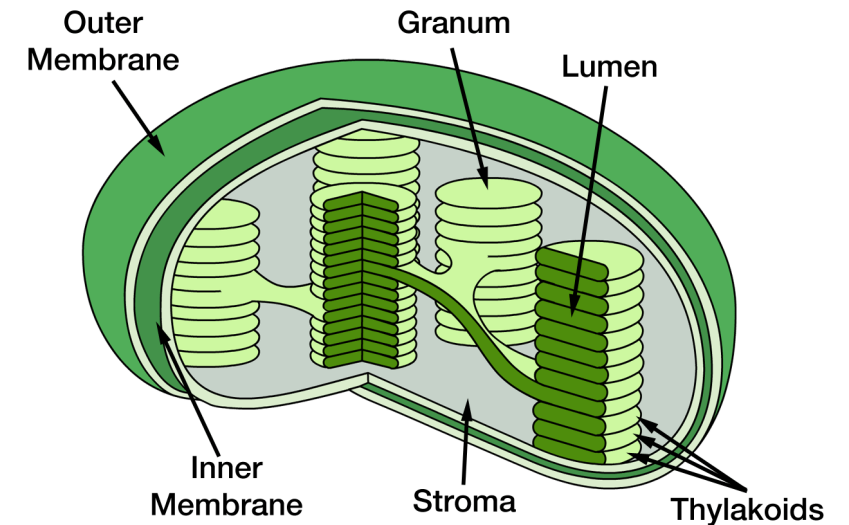


describe the structure of a dicotyledonous leaf, a palisade cell and a chloroplast and relate their structures to their roles in photosynthesis

Photosynthetic structures: the chloroplast

- The interior possesses a system of flattened membrane bound sacs called *thylakoids* which are stacked in other areas to form *grana* this provides a large surface area for the attachment of pigments, enzymes and electron carriers required for the light dependent reactions.
- The membrane of the grana contains the enzyme ATP synthase which uses energy released during electron transfer to synthesis ATP

Chloroplast



describe the relationship between structure and function in the chloroplast using diagrams and electron micrographs

Overview of photosynthesis

- Photosynthesis occurs in two stages
- Stage 1 is the light dependent reaction and it occurs on the membrane systems of the chloroplasts
- Stage 2 is the light independent reaction and it occurs in the stroma of the chloroplast

- *explain that energy transferred as ATP and reduced NADP from the light dependent stage is used during the light independent stage (Calvin cycle) of photosynthesis to produce complex organic molecules*
- *state the sites of the light dependent and the light independent stages in the chloroplast*

Light dependent reaction

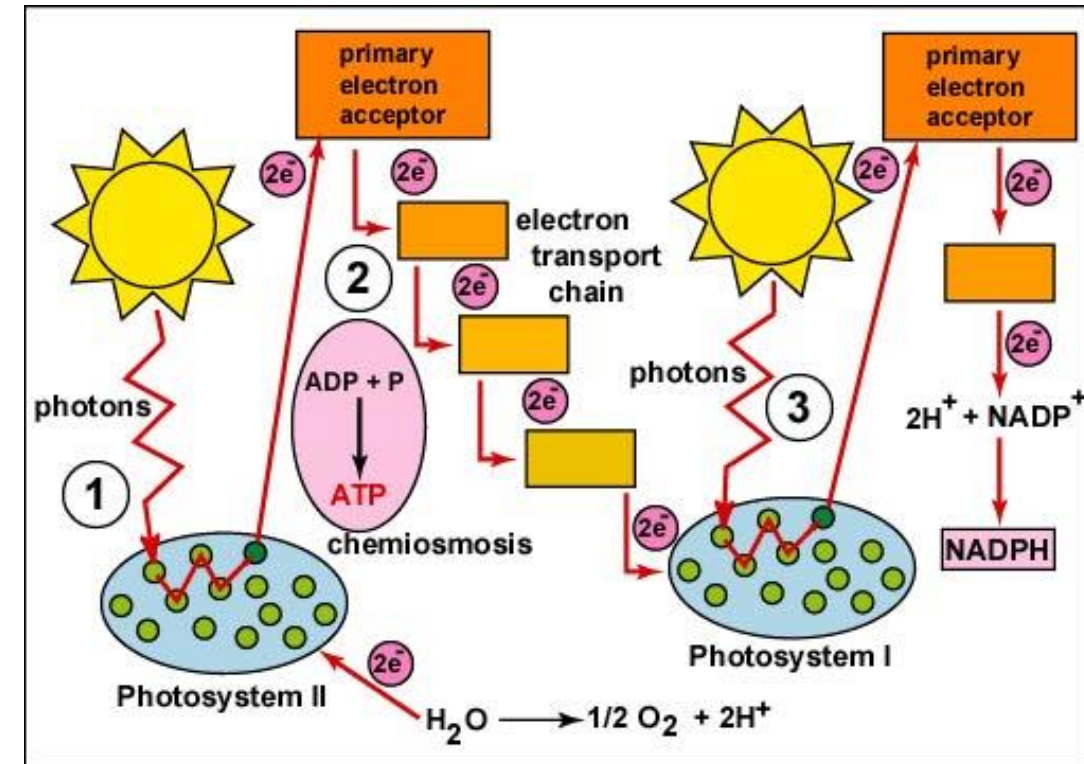
- **Site:** occurs on the grana and thylakoid membranes
- **Requirements:** chlorophyll and other pigments, NADP, water, ADP, Pi
- **Steps involved:** involves the photolysis of water to give protons, and the synthesis of ATP phosphorylation
- Role of pigments, interpret absorption and action spectra of pigments
- The light dependent reactions are facilitated by **photosystems**. Photosystems are two types: PSI and PSII.
- A photosystem is a complex assembly/collection of hundreds of accessory pigment molecules surrounding a primary pigment molecule. The light energy absorbed by the different accessory pigments is passed on to the primary pigment.

•explain that energy transferred as ATP and reduced NADP from the light dependent stage is used during the light independent stage (Calvin cycle) of photosynthesis to produce complex organic molecules

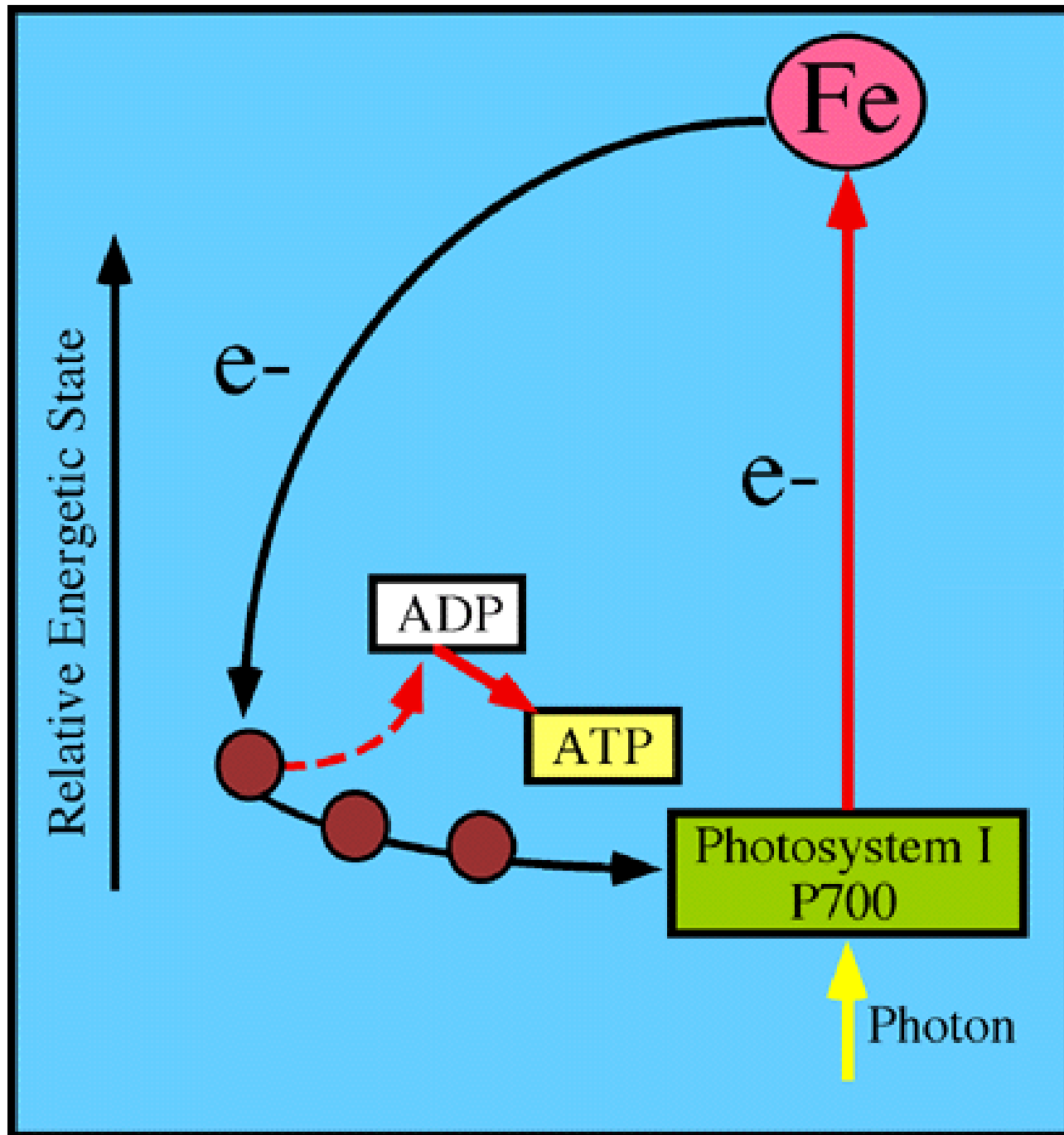
•describe the uses of ATP and reduced NADP in the light-independent stage of photosynthesis;

Cyclic photophosphorylation

- PS I
- *Photoactivation*: An electron in the chlorophyll molecule excited to a higher level and emitted
- The excited electron is captured by an electron acceptor and passed back to a chlorophyll molecule through a chain of electron carriers
- Energy released during this series of transfer is coupled into the synthesis of ATP (through chemiosmosis)

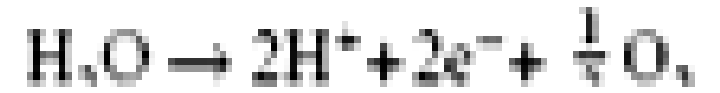


describe the light dependent stage as the photoactivation of chlorophyll resulting in the photolysis of water and the transfer of energy to ATP and reduced NADP (cyclic and non-cyclic photophosphorylation should be described in outline only)



Photolysis of water

- Enzymes in Photosystem II split water with light into H⁺ and e⁻
- $2\text{H}_2\text{O} \rightarrow 4\text{H}^+ + 4\text{e}^- + \text{O}_2$
- Some oxygen used for respiration
- Some diffuses out into the air

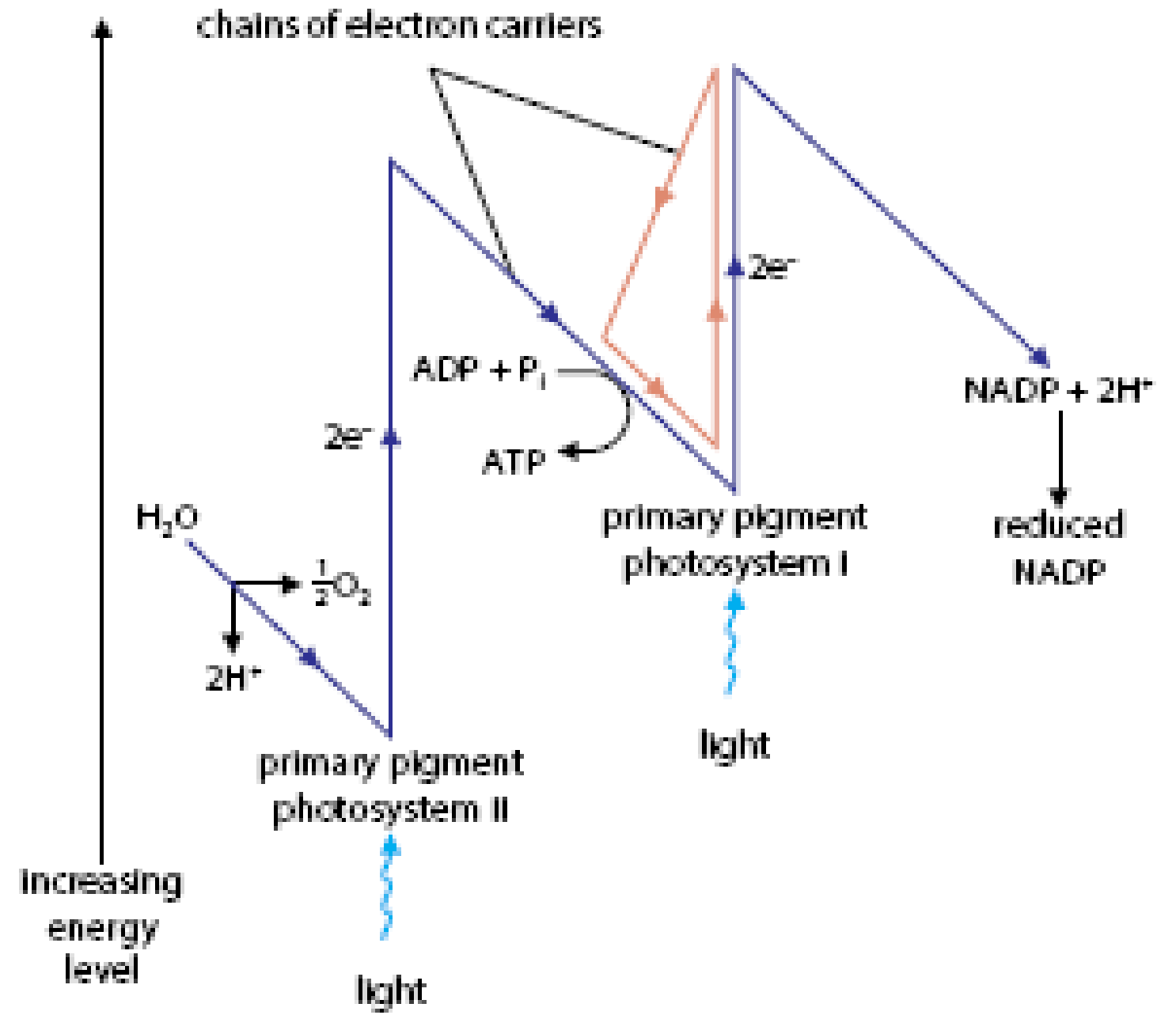


Non-cyclic photophosphorylation



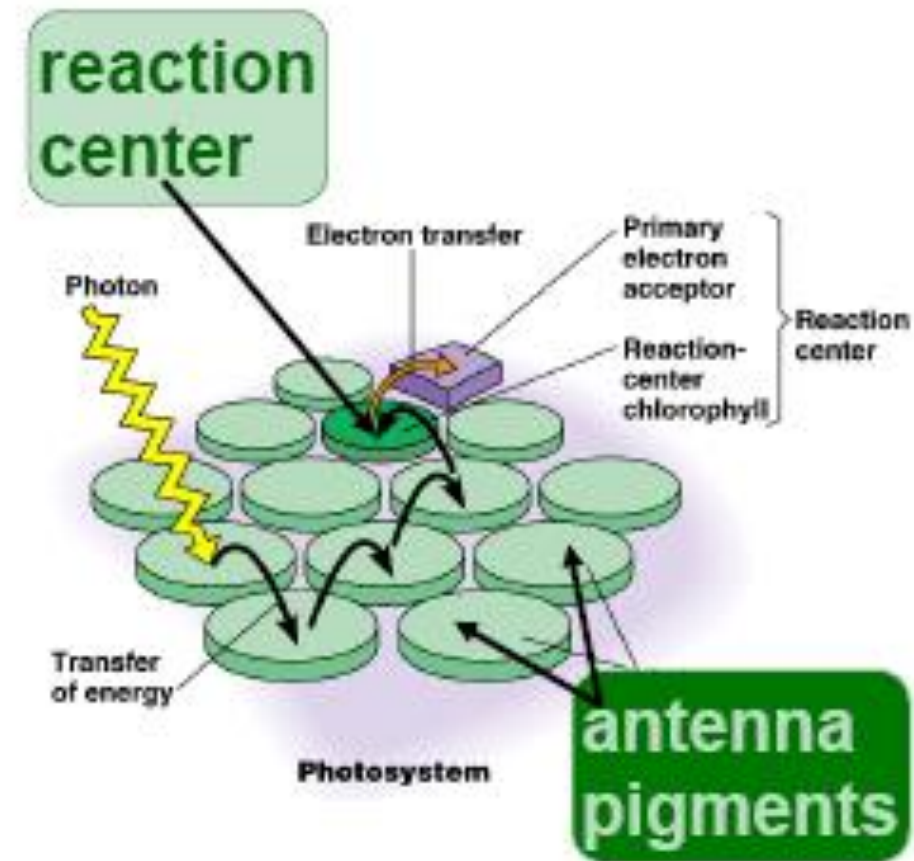
- Involves both PS I and PS II
- Light is absorbed by both photosystems and excited electrons are emitted from their reaction centres
- These electrons are absorbed by electron acceptors and transferred down a chain of electron carriers
- Light energy is also used to split water into hydrogen ions and two electrons and an atom of oxygen
- **PS I:** Electrons excited from PS I combine with the hydrogen ions produced during photolysis of water and are accepted by NADP to form reduced NADP . Then, PS I absorbs the electron excited from PS II
- **PS II:** Electrons excited from PS II are absorbed by PS I, and then, PS II receives its electron replacement from the electron released during the photolysis of water

describe the light dependent stage as the photoactivation of chlorophyll resulting in the photolysis of water and the transfer of energy to ATP and reduced NADP (cyclic and non-cyclic photophosphorylation should be described in outline only)



Role of pigments

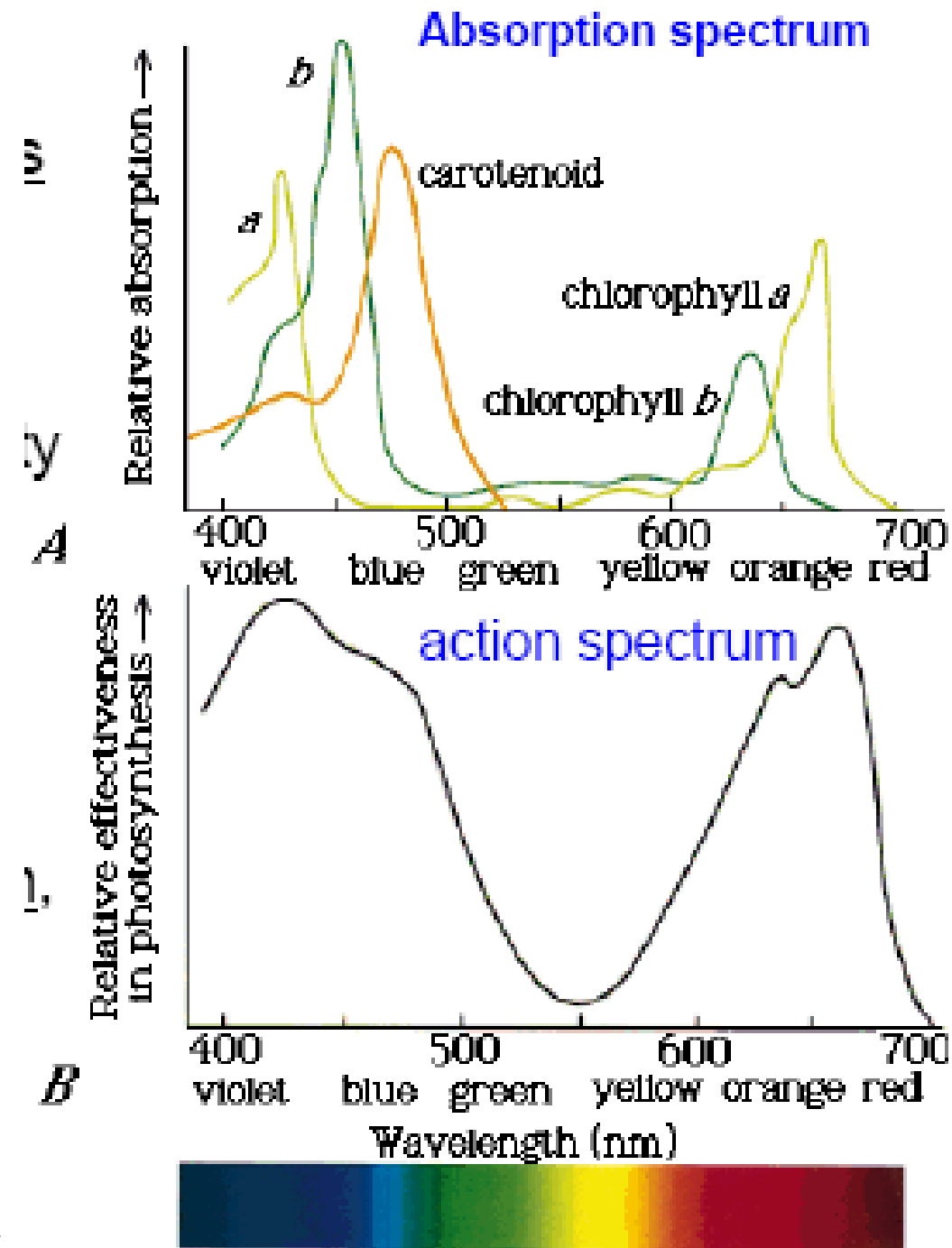
- Primary pigments: chlorophyll a and chlorophyll b
 - The pigments absorb light of different wavelength, but they absorb violet, blue and red best.
 - Most green wavelength is reflected back, thus leaves appear green
 - Chlorophyll a is the reaction centre of PS II, and it absorbs light at 680nm
 - Chlorophyll b is the reaction centre of PS I, and it absorbs light at 700nm
- Accessory pigments: carotene and xanthophyll
 - Carotenoids absorb light in violet, blue and green regions; they reflect yellow-orange



describe the role of chloroplast pigments (chlorophyll a, chlorophyll b, carotene and xanthophyll) in light absorption in the grana

Absorption and Action Spectra

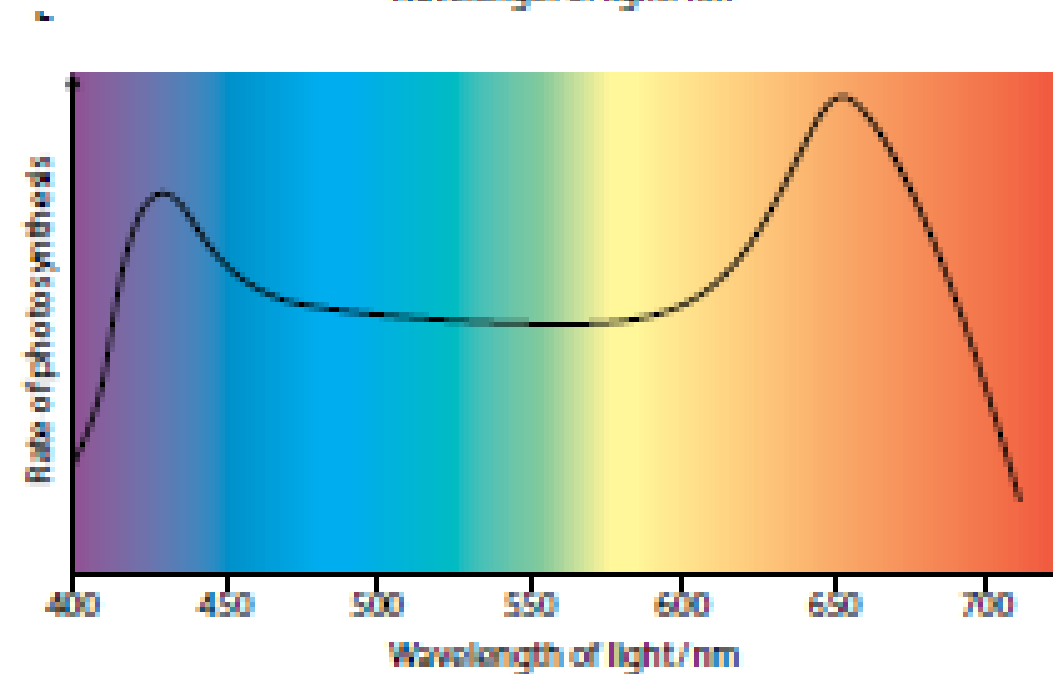
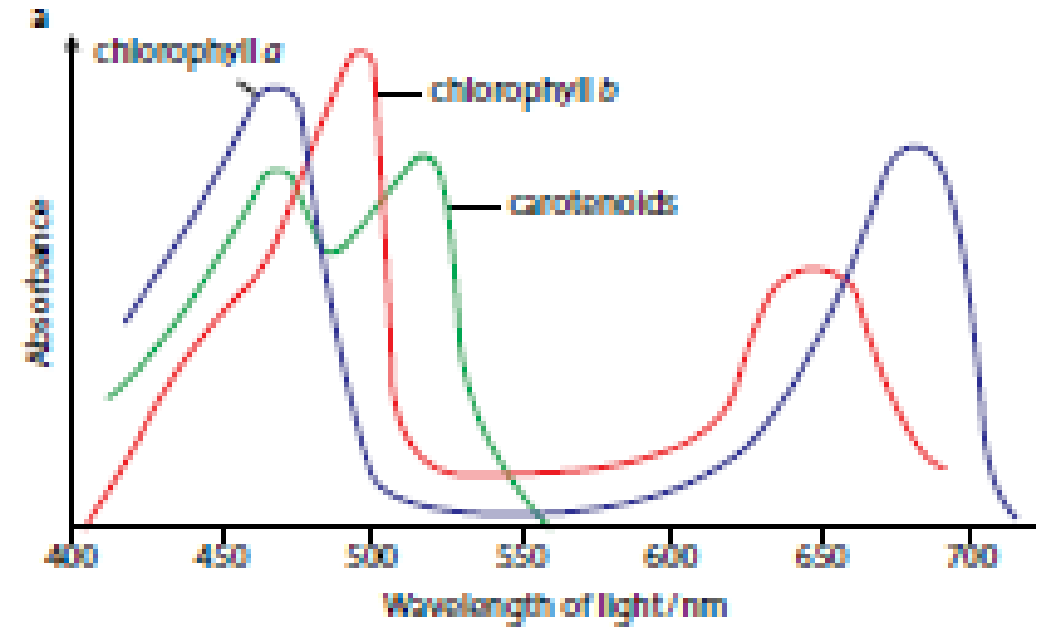
- **Absorption spectrum** is a graph that shows the absorbance of a pigment at different wavelength of light
- **Action spectrum** is a graph that shows the rate of photosynthesis at different wavelength of light



- *interpret absorption and action spectra of chloroplast pigments*
- *discuss the role of chloroplast pigments in absorption and action spectra, and separate them using chromatography*

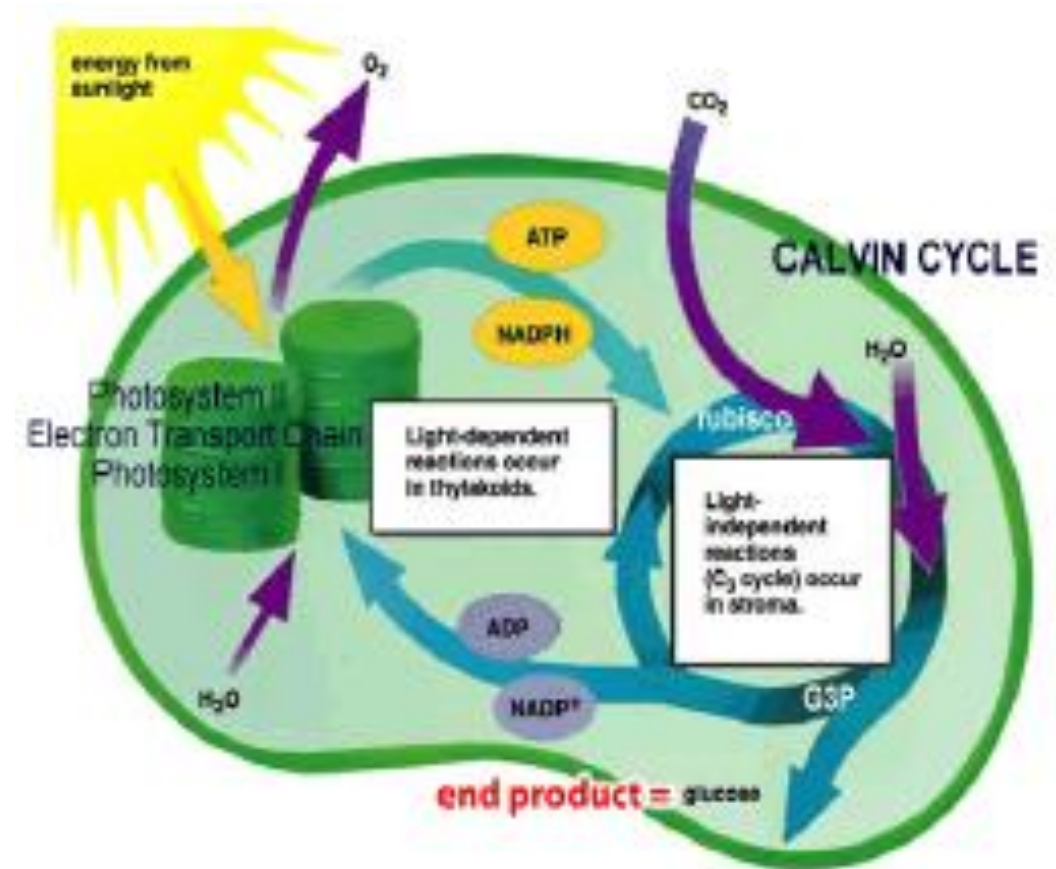
Absorption and action spectra

- The absorption spectra is a graph that shows the degree/quantity of light energy absorbed by various pigments in the chloroplast at different wavelength of light
- The action spectra is a graph that shows the rate of photosynthesis at different wavelength of light.
- The absorption spectra shows that **chlorophyll a** has an absorbance that increases as wavelength of light increases from 420 until it peaks at around 450 – 460nm after which it reduces sharply. The absorbance remains very low between 500nm – 580nm after which it begins to increase again and peaks at 680nm and then it reduces again as wavelength increases beyond 680nm. The peak at 450nm is greater than the peak at 680
- The absorption spectra of **chlorophyll b**...
- The absorption spectra of the **carotenoids**...
- The **action spectra** shows that...



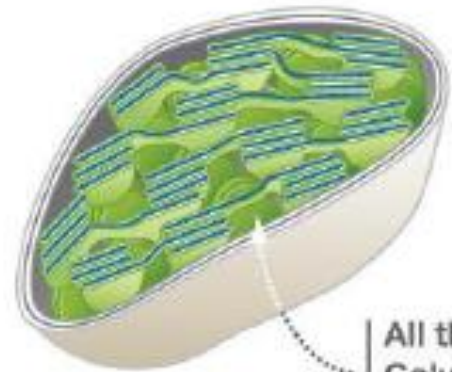
Calvin Cycle

- fixation of carbon dioxide by combination with ribulose biphosphate (RuBP), a 5C compound, to yield two molecules of GP (PGA), a 3C compound
- the reduction of GP to triose phosphate (TP) involving ATP and reduced NADP
 - the regeneration of ribulose biphosphate (RuBP) using ATP



outline the three main stages of the Calvin cycle:

(a) The Calvin cycle has three phases.



All three phases of the Calvin cycle take place in the stroma of chloroplasts

1. Fixation



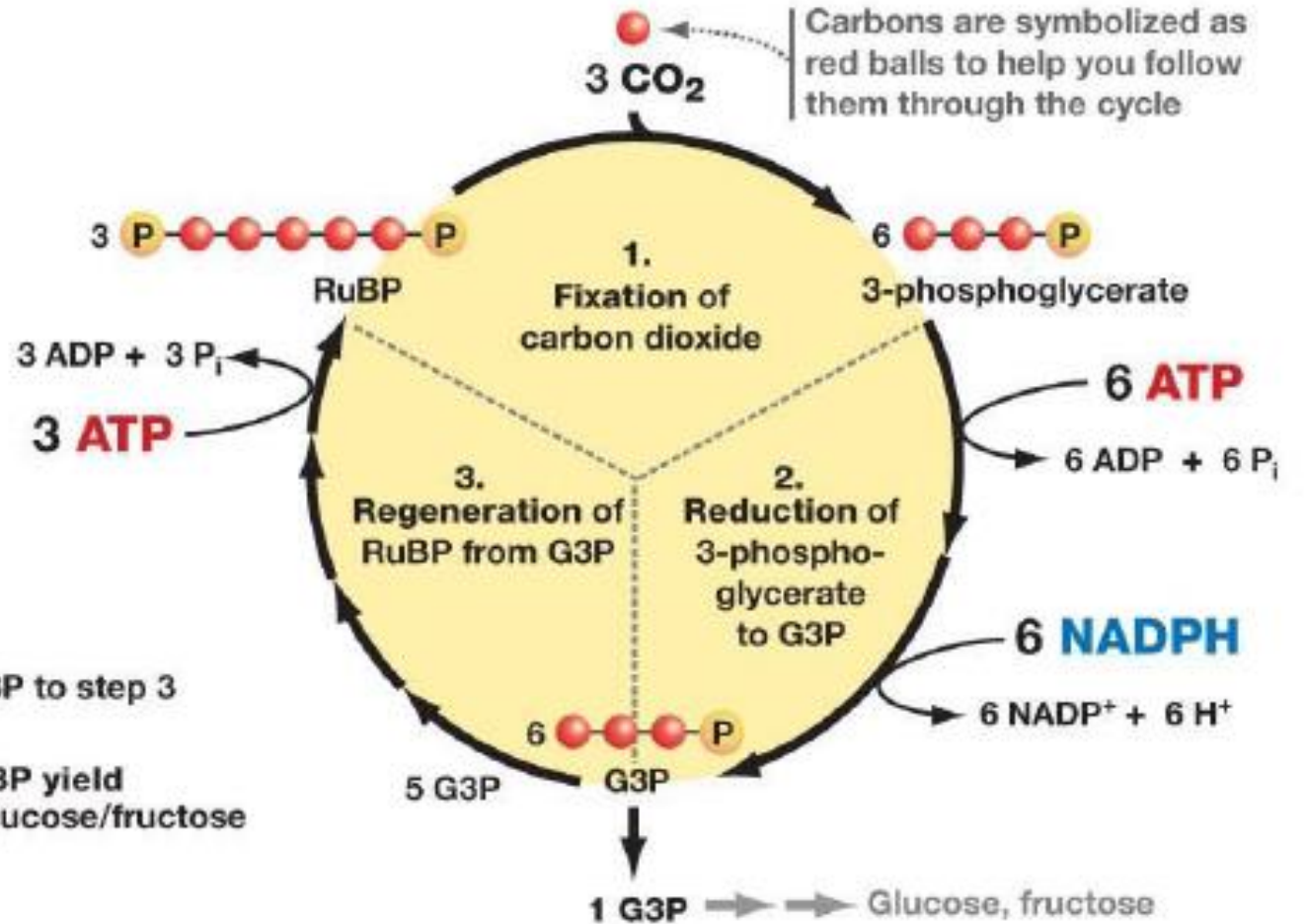
2. Reduction



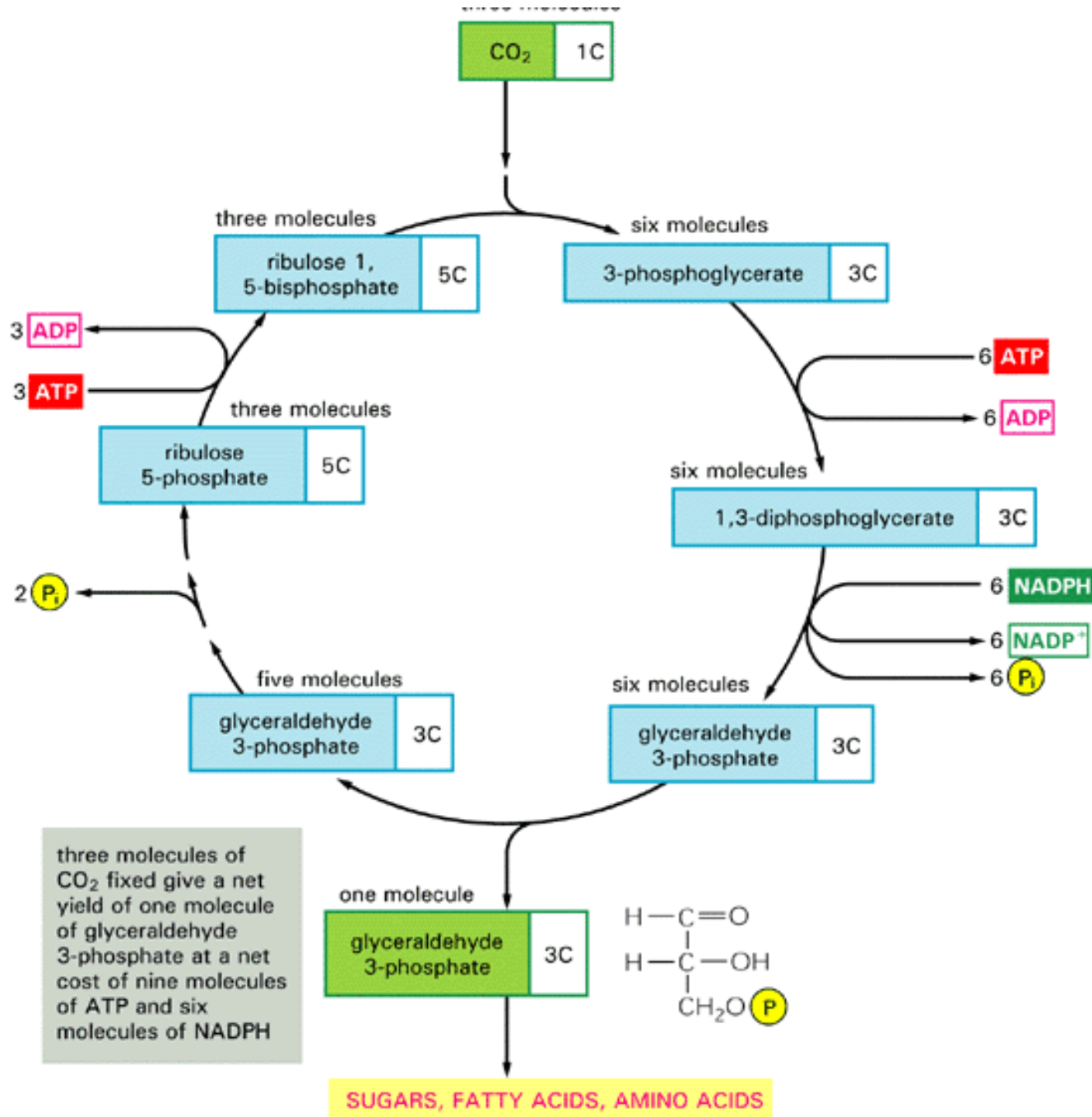
3. Regeneration



(b) The reaction occurs in a cycle.



Conversion of Calvin Cycle intermediates



- Carbon dioxide diffuses into leaf through stomata on underside of leaf and enters stomata
- Combines with 5-carbon ribulose bisphosphate (RuBP) in a reaction catalysed by the enzyme *rubisco*
- Two 3-carbon glycerate 3-phosphates (GP) created
- GP is reduced and phosphorylated by NADP and ATP to make triose phosphate (TP)
- Triose phosphate goes on to make sugars e.g. glucose
- RuBP is reformed when TP combines with ATP

describe, in outline, the conversion of Calvin cycle intermediates to carbohydrates, lipids and amino acids and their uses in the plant cell

Conversion of Calvin cycle intermediates

- Some of the triose phosphate produced at the end of Calvin Cycle will be used to regenerate RuBP
- Some will condense to form hexose phosphate which will then be used to produce starch, sucrose or cellulose
- Some will be converted to glycerol and fatty acid which are used to produce lipids for cell membrane synthesis
- Some will be converted to acetyl coenzyme A and be used for respiration
- Some will be used for synthesis of amino acids

describe, in outline, the conversion of Calvin cycle intermediates to carbohydrates, lipids and amino acids and their uses in the plant cell

Overview of photosynthesis

Light reactions

produced **ATP**

produced **NADPH**

consumed **H₂O**

produced **O₂** as byproduct

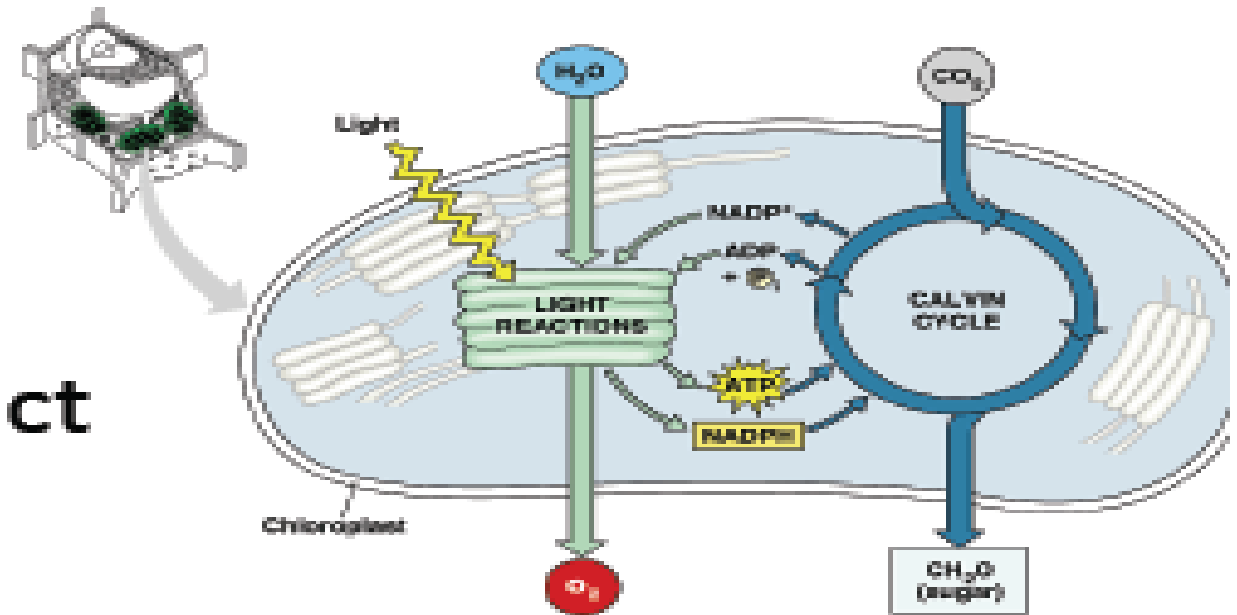
Calvin cycle

consumed **CO₂**

produced **G3P (sugar)**

regenerated **ADP**

regenerated **NADP**



Limiting Factors

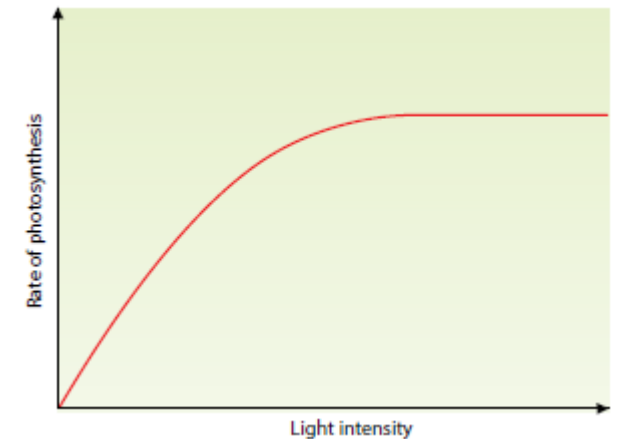
- A limiting factor is one of the many factors that affect rate of photosynthesis when it is near its lowest value and every other factor is at its optimum.
- The rate of photosynthesis is affected by light intensity, carbon dioxide concentration and temperature; and all three factors work together to influence the rate.
- At every point in time, one of those three factors is always the *limiting factor*. This means that when the value of the other two factors are changed, the rate of photosynthesis will not be altered (since they are not the limiting factor). But when the value of limiting factor is changed, the rate of photosynthesis will be altered.

explain the term limiting factor in relation to photosynthesis

explain the effects of changes in light intensity, carbon dioxide concentration and temperature on the rate of photosynthesis

Effect of light intensity

- As light intensity increases, the rate of photosynthesis increases
- Light is essential for the light dependent stage of photosynthesis
- The greater the light intensity, the greater the rate of formation of ATP during photophosphorylation (cyclic and non cyclic), the greater the rate of formation of reduced NADP.
- And more of these are available for the light independent reactions
- However at higher light intensity, the rate no longer increases, because at such point, light ceases to be a limiting factor



explain the term limiting factor in relation to photosynthesis

explain the effects of changes in light intensity, carbon dioxide concentration and temperature on the rate of photosynthesis

Effect of CO₂ Concentration

- As **CO₂ Concentration** increases, the rate of photosynthesis increases, and this continues until **CO₂ Concentration** ceases to act as the limiting factor.
- This is because **CO₂** is required for the light independent stage of photosynthesis.
- The greater the concentration of **CO₂**, the faster the rate of activity of the enzymes that are involved in the Calving Cycle, e.g. enzymes such as RUBISCO

explain the term limiting factor in relation to photosynthesis

explain the effects of changes in light intensity, carbon dioxide concentration and temperature on the rate of photosynthesis

Effect of Temperature + light intensity

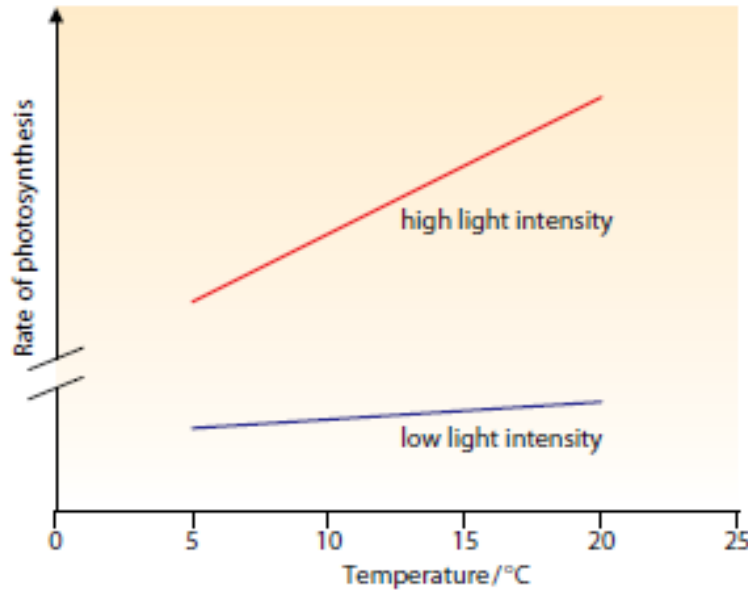


Figure 13.8 The rate of photosynthesis at different temperatures and constant light intensities.

- Increasing temperature increases the rate of photosynthesis because increase in temperature increases the kinetic energy of the enzymes that catalyse the different reactions occurring during the light dependent (e.g. water splitting enzyme) and light independent (e.g. RUBISCO) reaction
- However, this is also not indefinite because once the optimum temperature of these enzymes are exceeded, the enzymes become denatured.
- From the graph, results show that at low light intensity, the rate photosynthesis increases with temperature but not as fast as it increases at high light intensity.
- This is simply because light is essential for production of ATP and reduced NADP which will be used for the light independent reaction that involves lots of enzymes.

explain the term limiting factor in relation to photosynthesis

explain the effects of changes in light intensity, carbon dioxide concentration and temperature on the rate of photosynthesis

Limiting factors: applications

- The knowledge of how limiting factors affect the rate of photosynthesis has been applied by farmers to increase the yield of their farm produce.
- They can construct glasshouses and create a protected and controlled environment where the **temperature, CO₂ concentration and light intensity** are optimised for optimal rate of photosynthesis.
- If the rate of photosynthesis is as high as possible, the quantity of food production by the plants will be increased, and thus the farm yield will be increased.

explain how an understanding of limiting factors is used to increase crop yields in protected environments, such as glasshouses

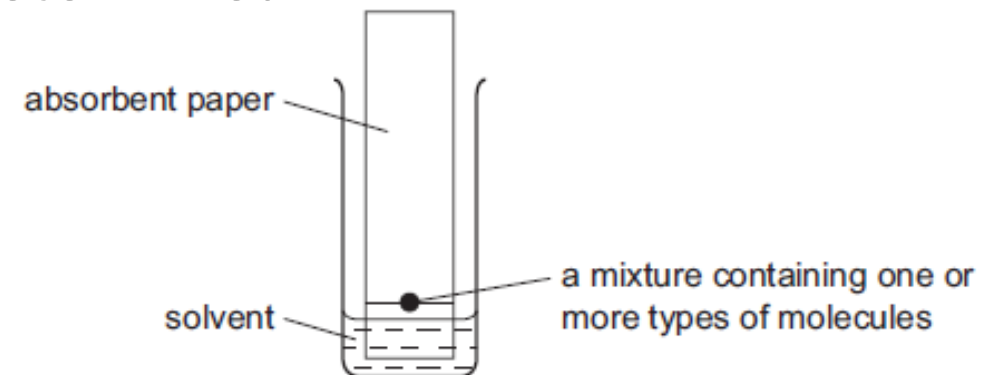
Experiment 1: Chromatography

- To separate the various pigments that are found within a leaf using chromatography, the following steps are followed:
- The leaves are plucked from the plant and allowed to dry. They are then ground with a suitable solvent, e.g. propanone. Once grounded with the solvent, various pigment substances within the leaf will blend into the solvent to form a leaf extract. This leaf extract contains mixture of pigments. This leaf extract is allowed to concentrate by putting the mixture in a dryer which will evaporate a large portion of the solvent.
- A chromatography paper containing the stationary phase is prepared and a pencil line is drawn around the bottom of the paper to indicate the point of origin.
- The extract is then placed on this pencil line by repetitive spotting
- The paper is then placed (vertically) in jar of (different) solvent, ensuring that the solvent in side the jar does not cover the line of origin and the solvents that have been spotted onto it.

use chromatography to separate and identify chloroplast pigments and carry out an investigation to compare the chloroplast pigments in different plants (reference should be made to *Rf values in identification*)

- The solvent in the jar will then begin to rise of the paper and as it rises, it will mix with the components of the leaf extract and separate the pigments causing each pigment to travels up the paper at different speed ;
- As the pigments they, they are separated from each other, and at the end, they will arrive at different positions on the chromatography paper. The distance moved by each pigment is unique and this distance can be used to calculate the R_f of that pigment value ;
- The process can be carried out along two dimensions (two dimensional chromatography) so as to ensure better separation of the pigments ;
- The measure distance travelled by solvent (front) and pigment (spot), and this distance is used to calculate the R_f value using the formula below:
- Once the R_f value is calculated, it can be compared with known R_f values so that the identity of the pigment can be determined.

(calculate) R_f value = $\frac{\text{distance travelled by pigment}}{\text{distance travelled by solvent (front)}} ;$



Experiment 2: DCPIP

carry out an investigation to determine the effect of light intensity or light wavelength on the rate of photosynthesis using a redox indicator (e.g. DCPIP) and a suspension of chloroplasts (the Hill reaction)

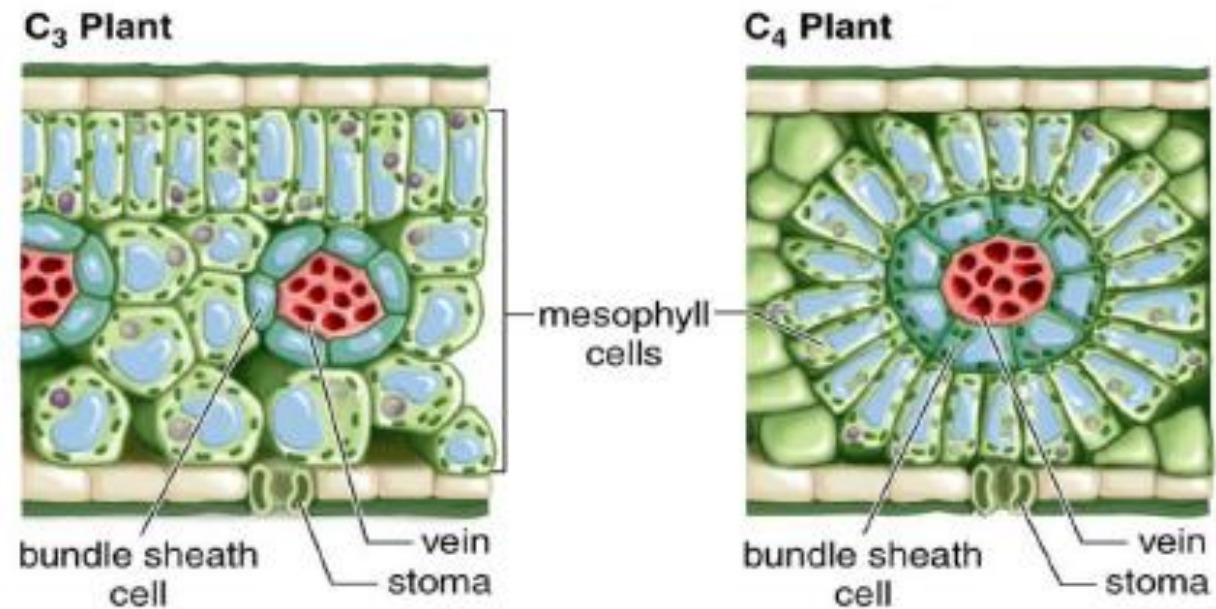
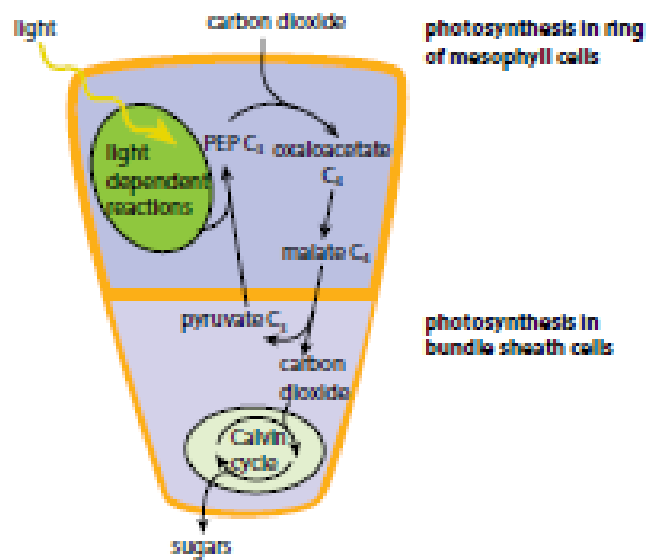
Experiment 3: Limiting factors

carry out investigations on the effects of light intensity, carbon dioxide and temperature on the rate of photosynthesis using whole plants, e.g. aquatic plants such as *Elodea* and *Cabomba*

Adaptations of C4 plants

- C4 plants are plants that are adapted to prevent the process of ***photorespiration*** from taking place in the leaves of such plants.
- What is photorespiration? This is a reaction that involves the reaction of RuBP with oxygen (catalysed by rubisco), as a result of high environmental temperature and high light intensity. This reaction reduces the quantity of RuBP available to bind with CO₂ during the first step of Calvin Cycle. Thus results in reduced rate of photosynthesis.
- The spatial separation of initial carbon fixation from the light dependent stage (biochemical details of the C4 pathway are required in outline only)
- The high optimum temperatures of the enzymes involved

explain how the anatomy and physiology of the leaves of C4 plants, such as maize or sorghum, are adapted for high rates of carbon fixation at high temperatures in terms of:



- These C₄ plants are common in tropical areas of the world (where conditions of high temperature and high light intensity is prevalent) e.g. are maize, sorghum, and sugar cane.
- These plants have the following features (/how photosynthesis occurs in C₄ plants).
 - The cells that contain RuBP and rubisco (called Bundle Sheath cells) are arranged around the vascular bundles of the leaf. They are then surrounded by another ring/layer of mesophyll cells such that air inside the leaf cannot reach the bundle sheath cells
 - This layer of mesophyll cells that surround the bundle sheath cells are in direct contact with air in the leaf and they contain an enzyme called PEP Carboxylase (Phosphoenol pyruvate). PEP carboxylase catalyses the conversion of PEP with CO₂ to form a 4c compound called oxaloacetate
 - Oxaloacetate is then converted to malate (inside these mesophyll cells) and the malate is transported into the bundle sheath cells. The malate is decarboxylated, its CO₂ is released to combine with RuBP to form GP and continue the process of Calvin Cycle.
 - Generally enzymes in C₄ plants have higher optimum temperature compared to enzymes in C₃ plants, and as a result, they have higher rate of photosynthesis.