PLANT PHYSIOLOGY
Photosynthesis: C4 and CAM Plants

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MODUL 05

SELF-PROPAGATING ENTREPRENEURIAL EDUCATION DEVELOPMENT

Tanaman C4 dan CAM adalah tanaman yang menggunakan lintasan fixasi CO_2 tertentu untuk meningkatkan konentrasi CO_2 di sekitar RUBISCO dalam khloroplast. Ini berbeda dari tanaman C3 yang hanya menggunakan siklus Calvin yang juga terdapat pada tanaman C4 dan CAM.

LECTURE OUTCOMES

After the completion of this lecture and mastering the lecture materials, students should be able;

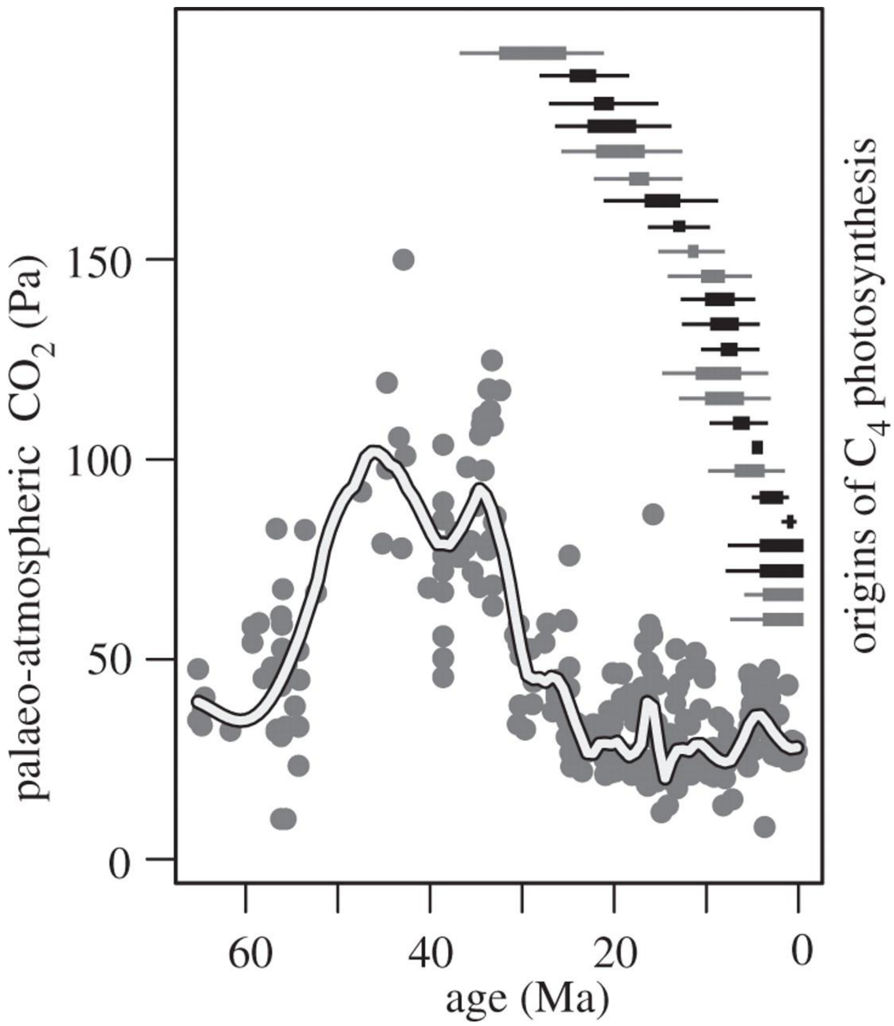
1. To explain the assimilation of CO_2 to be carbohydrate (sugars) in C4 and CAM plants
2. To explain the diffusion of CO_2 from the atmosphere into the site of assimilation in the chloroplasts of C4 and CAM plants
3. To explain reactions, enzymes and products involved in the reduction of CO_2 to be carbohydrate in C4 and CAM plants

1. C4 Plants	2. CAM Plants
C4 Plant Evolution	CAM Plant Evolution
Discovery C4 Pathway	CAM Plant Characteristics
Leaf Anatomy of C4 plants	CO_2 Reduction
CO_2 Reduction	
Type of C4 Plants	
Energetic of the C4 Photosynthetic System	

1. C4 PLANTS

1. C4 Plant Evolution

1. C4 photosynthesis has evolved more than 60 times as a carbon-concentrating mechanism to augment the ancestral C3 photosynthetic pathway.
2. C_4 origins have all occurred over the past 30 Myr, with no difference in timing between monocot and eudicot lineages.
3. It is hypothesized that **atmospheric CO_2 depletion** coupled with high **temperatures**, **open habitat** and **seasonally dry subtropical environments** caused **excessive demand for water transport**, and **selected for C4 photosynthesis** to enable lower stomatal conductance as a water-conserving mechanism.



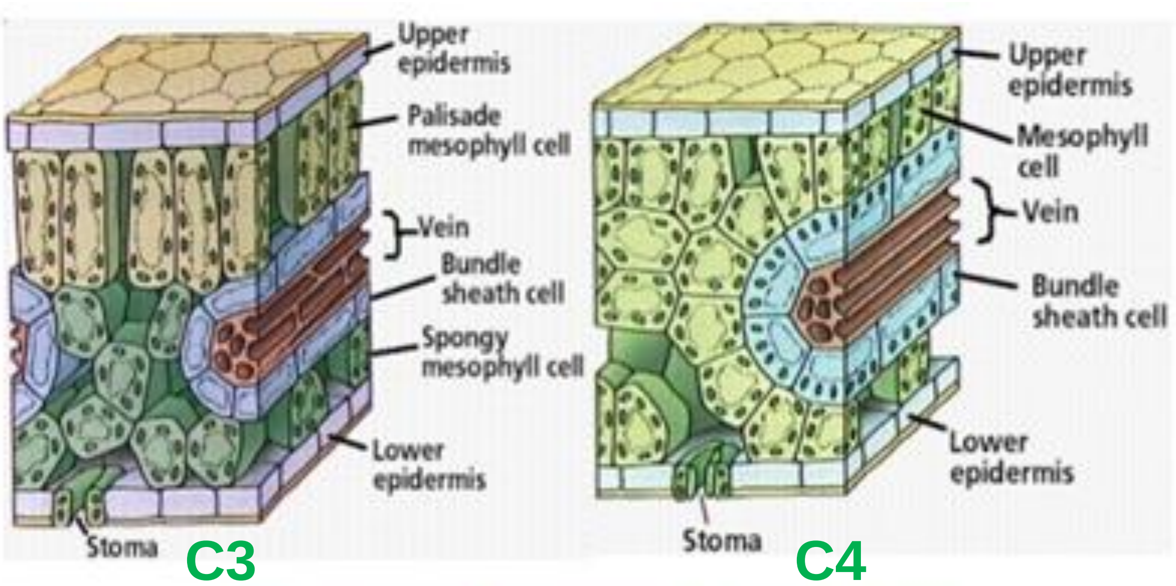
Geological history of atmospheric CO_2 and the estimated ages of C4 evolutionary origins. Geological history of atmospheric CO_2 and the estimated ages of C_4 evolutionary origins. The estimated ages of C_4 evolutionary origins in grasses (dark grey horizontal bars) and eudicots (black horizontal bars) were obtained using phylogenetic inference and calibration to fossils [35]. Thick bars represent uncertainty in the position of each C_4 evolutionary origin on the phylogeny, while thin bars indicate uncertainty in dating of the phylogeny (reproduced with permission from Christin *et al*. [35]). The palaeo-atmospheric CO_2 history of the Cenozoic is reconstructed from multiple independent proxies (pale grey circles), with a smoothed line of best fit encompassing all of the evidence (reproduced with permission from Beerling & Royer [4]).
Colin P. Osborne, and Lawren Sack Phil. Trans. R. Soc. B 2012;367:583-600

2. Discovery C4 Pathway

1. In the late 1950s, H. P. Kortschack and Y. Karpilov observed early labeling of 4-carbon acids when “CO_2 was provided to sugarcane and maize.
2. After leaves were exposed to “CO_2 for a few seconds in the light, 70 to 80% of the label was found in the 4-carbon acids **malate** and **aspartate**—a pattern very different from the one observed in leaves that photosynthesize solely via the Calvin—Benson cycle.
3. M. D. Hatch and C. R. Slack elucidated C4 cycle, and established that malate and aspartate are the first stable, detectable intermediates of photosynthesis in leaves of sugarcane.
4. The carbon 4 of malate subsequently becomes carbon 1 of 3-phosphoglycerate (Hatch and Slack 1966).

3. Leaf Anatomy of C4 Plants

1. The key features of the C4 cycle are the presence of two distinctive photosynthetic cell types: an internal ring of bundle sheath cells where RUBISCO is located, which is wrapped with an outer ring of mesophyll cells.
2. The chloroplasts in bundle sheath cells are concentrically arranged and exhibit large starch granules and unstacked thylakoid membranes.
3. On the other hand, mesophyll cells contain randomly arranged chloroplasts with stacked thylakoids and little or no starch.
4. However, there are now clear examples of single-cell C4 photosynthesis in a number of green algae, diatoms, and aquatic and land plants (Edwards *et al*. 2004; Muhaidat *et al*. 2007) (Fig. 8.12A).



Comparison between C_3 and C_4 Leaf

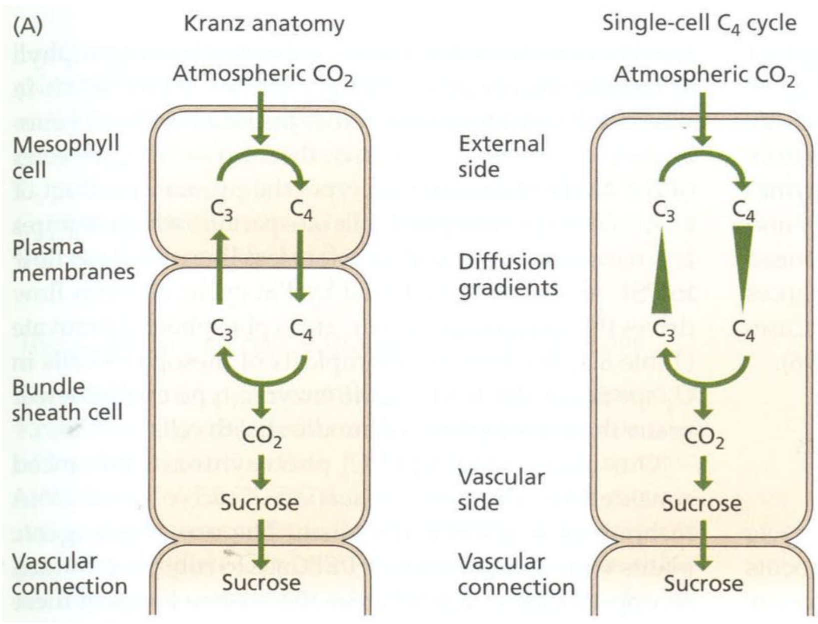


Fig. 8.12 The photosynthetic pathway in leaves. (A) In almost all known C4 species, photosynthetic CO_2 assimilation requires the development of Kranz anatomy (left panel). A few land plants, typified by *Borszczowia aralocaspica* and Bienertia cycloptera, contain the equivalents of the C4 compartmentalization in a single cell (right panel).

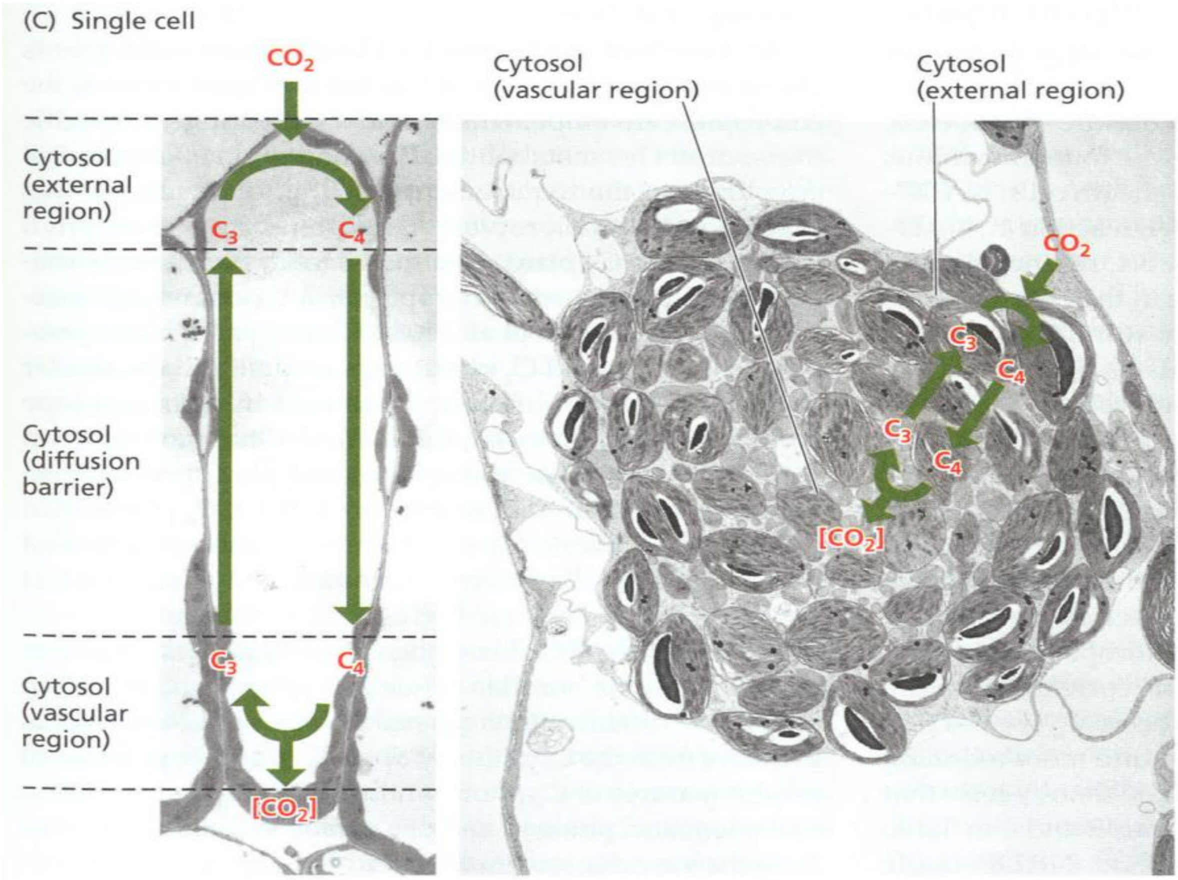
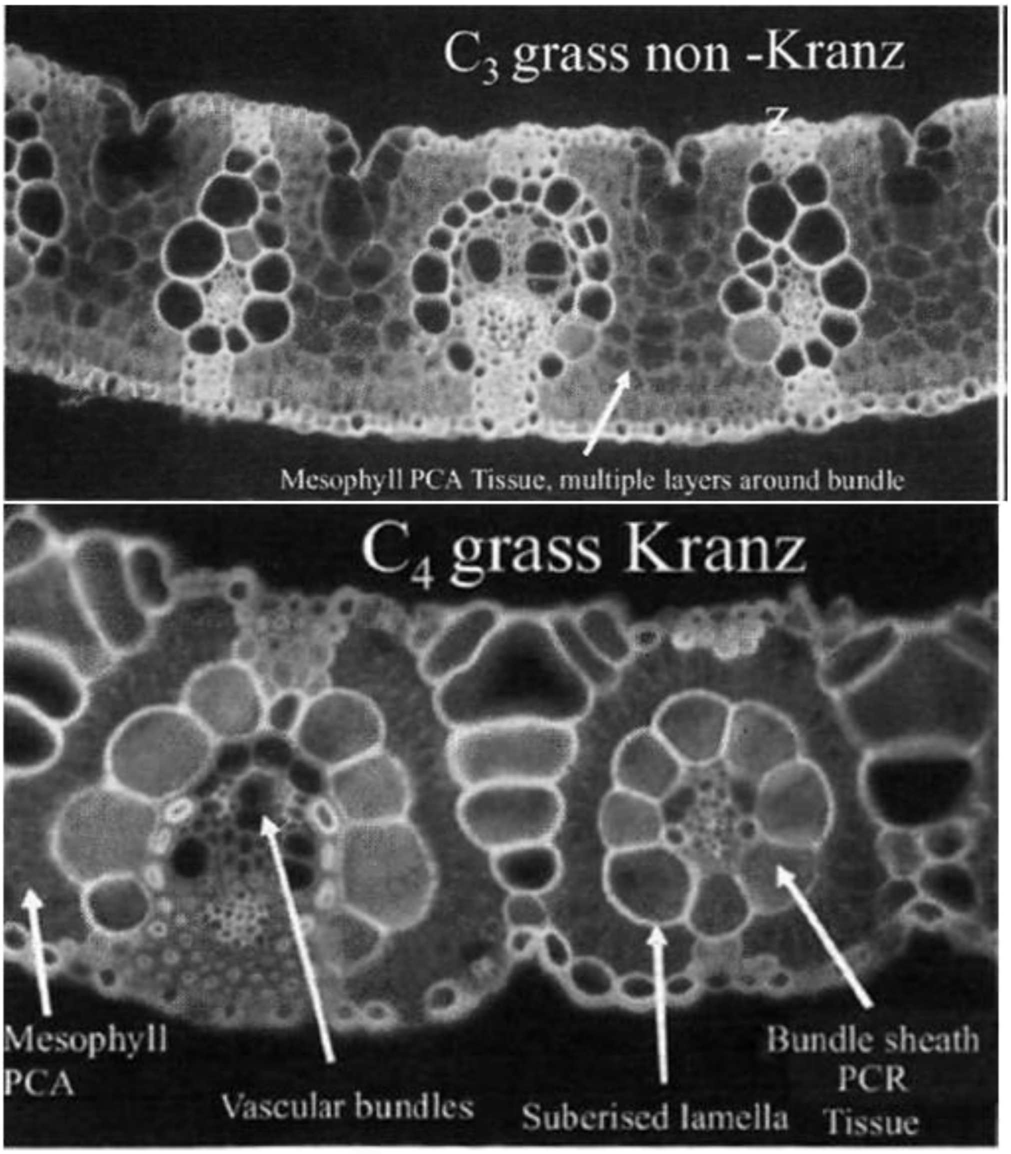
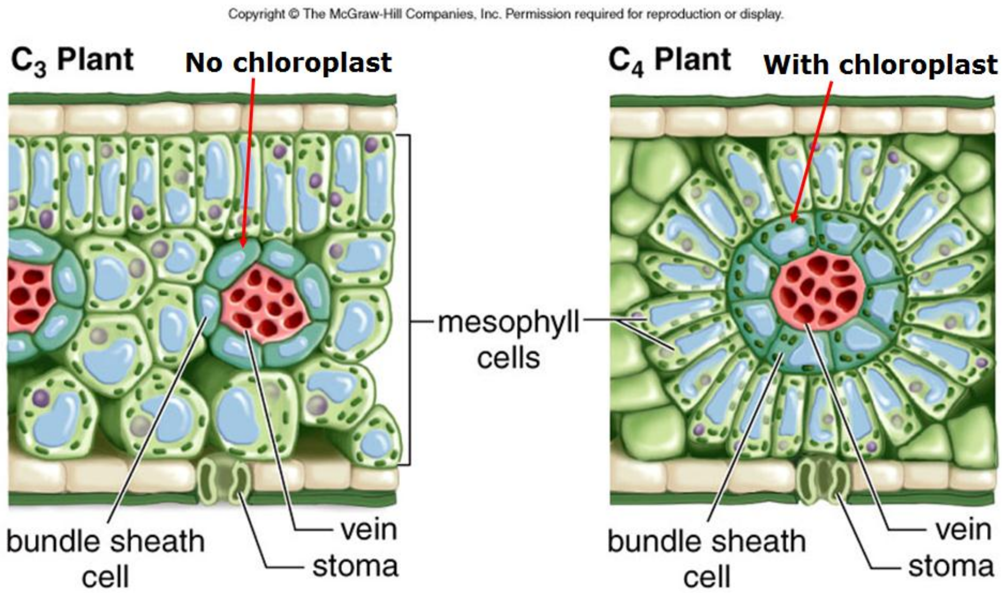


Fig. 12.8C. Single-cell C4 photosynthesis. Diagrams of the C4 cycle are superimposed on electron micrographs of *Borszczowia aralocaspica* (left) and *Bienertia cycloptera* (right). (B courtesy of Athena McKown; C from Edwards *et al*. 2004.)



Comparison between leaf structure of C3 plants and C4 plants under lectron microscope



Leaf structure model of C3 plants (leaft) and C4 plants (right)

4. CO_2 Reduction

1. The transport of CO_2 from the external atmosphere to the bundle sheath cells proceeds through five successive stages (Fig. 8.11).
2. In the C4 cycle, the enzyme phosphoenolpyruvate carboxylase (PEPCase), rather than rubisco, catalyzes the primary carboxylation, the reaction of HCO_3^- with PEP (phosphoenolpyruvate) (Sage 2004).
3. The 4-carbon reaction product, **oxaloacetate**, is converted into **malate** or **aspartate** (depending on the species) by NADP-malate dehydrogenase or aspartate aminotransferase, respectively.
4. Malate or aspartate is exported to bundle sheath cells where it is decarboxylated, releasing CO_2 that is refixed by rubisco via the Calvin cycle.
5. The specific paths by which CO_2 is concentrated in the vicinity of rubisco vary substantially between different C4 species.

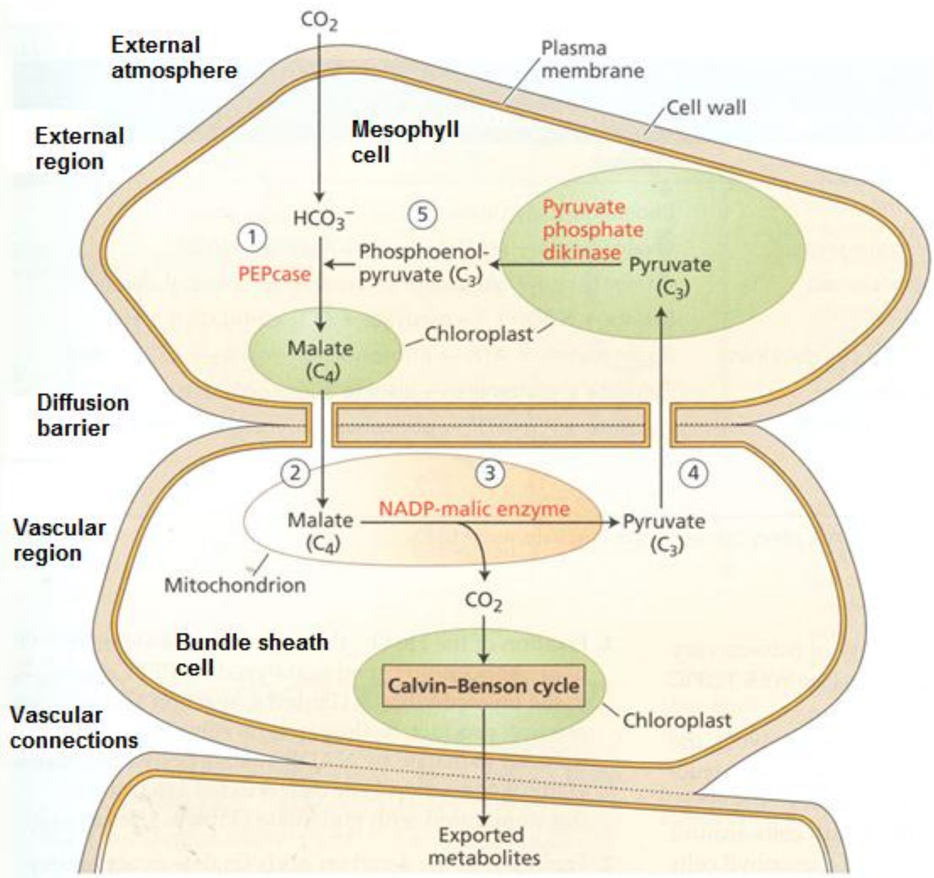
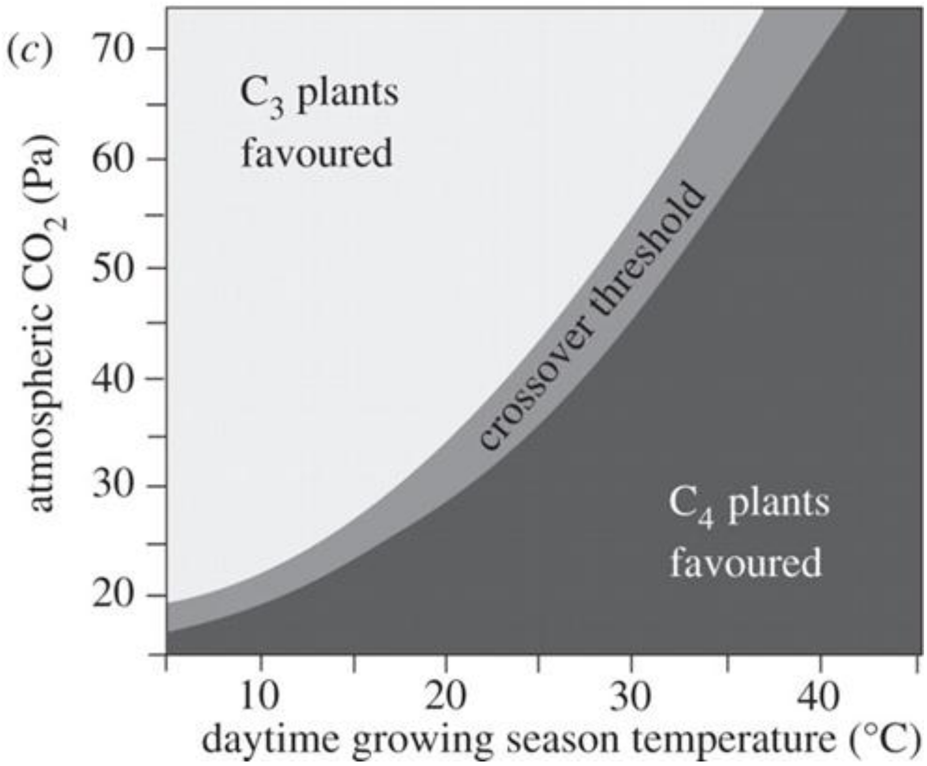


Fig. 8.11 The C4 photosynthetic carbon cycle involves five successive stages in two different compartments



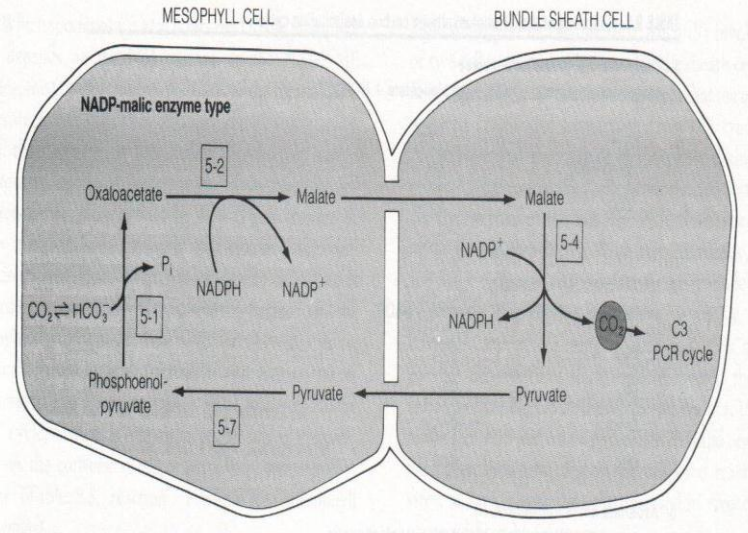
Condition favoured by C3 plants and C4 plants. Osborne & Sack (2012)

5. Types of C4 Plants

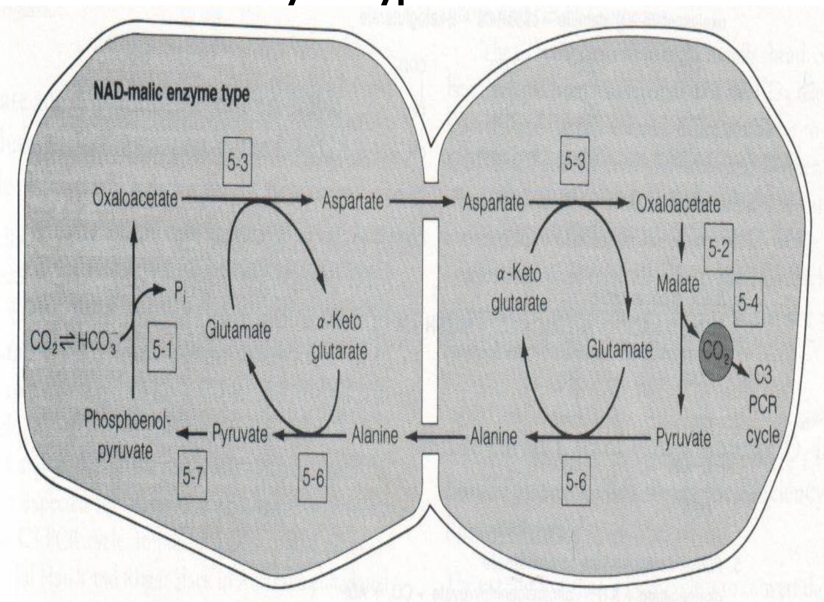
Principal C4 acid transported to the BSC	Decarboxylating enzyme	Variant name	Principal C3 acid returned to MC	Examples
Malate	NADP-dependent malic enzyme (chloroplast)	NADP-ME	Pyruvate	Maize, crabgrass, sugarcane, sorghum
Aspartate	NAD-dependent malic enzyme (mitochondria)	NAD-ME	Alanine	Millet, Pigweed (Panicum miliaceum)
Aspartate	Phosphoenolpyruvate carboxykinase	PEP-CK	Alanine/ pyruvate	Guinea grass (Panicum maximum)

BSC = bundle sheath cells & MC, mesophyll cells

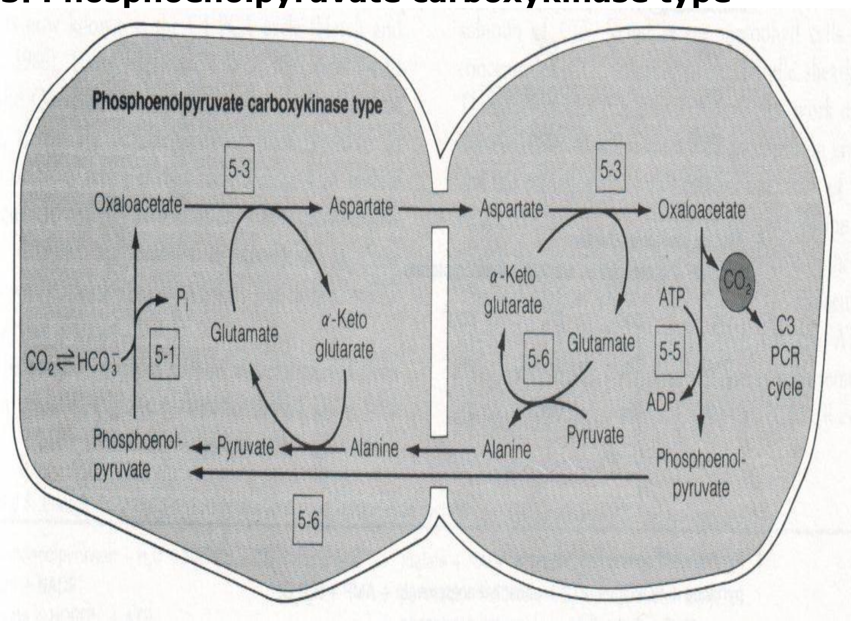
1. NADP-malic enzyme type



2. NAD-malic enzyme type



3. Phosphoenolpyruvate carboxykinase type



6. Energetics of the C4 Photosynthetic System

$$\text{phosphoenolpyruvate} + H_2O + NADPH + CO_2\ (\text{mesophyll}) \rightarrow \text{malate} + NADP^+ + HOPO_3^{2-}$$
$$\text{malate} + NADP^+ \rightarrow \text{pyruvate} + NADPH + CO_2\ (\text{bundle sheath cell})$$
$$\text{pyruvate} + HOPO_3^{2-} + ATP \rightarrow \text{phosphoenolpyruvate} + AMP + 2H^+ + P_2O_7^{4-}$$
$$P_2O_7^{4-} + H_2O \rightarrow 2HOPO_3^{2-}$$
$$AMP + ATP \rightarrow 2ADP$$
$$\text{Net:}\quad CO_2\ (\text{mesophyll}) + 2ATP + 2H_2O \rightarrow CO_2\ (\text{bundle sheath cell}) + 2ADP + 2HOPO_3^{2-} + 2H^+$$

Cost of concentrating CO_2 within bundle sheath cell = **2ATP per CO_2**

The reduction cost of 1 mol CO_2 via PCR = **2mol NADPH+3 mol ATP**

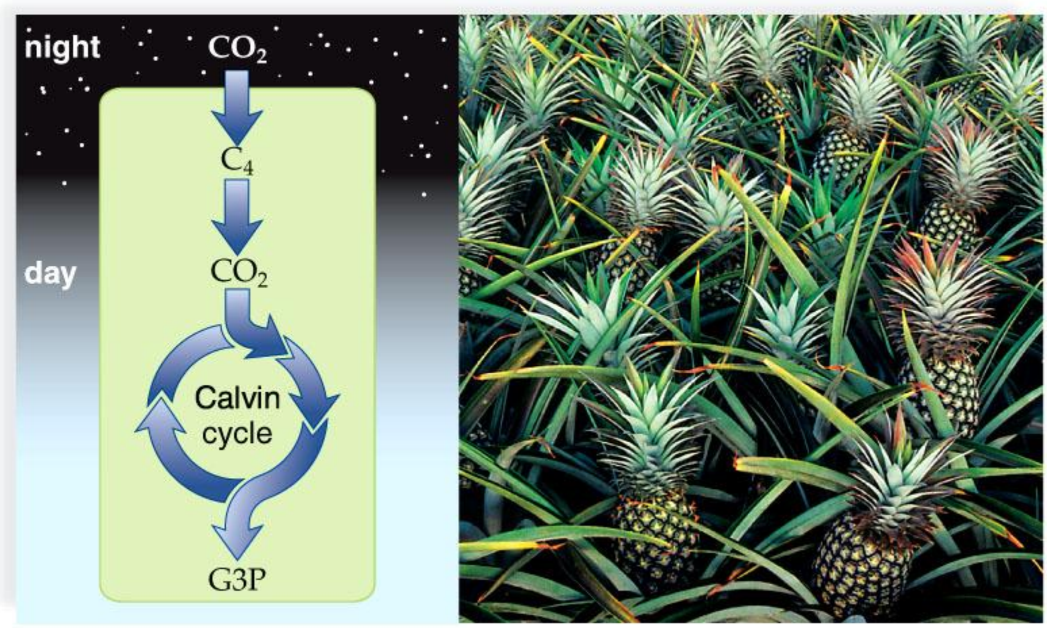
Total reduction cost of 1 mol CO_2 in C4 plants =?

2. CAM PLANTS

1. CAM Plant Evolution

1. Many plants that inhabit arid environments with seasonal water availability such as pineapple (*Ananas comosus*), agave (*Agave* spp.), cacti (*Cactaceae*), and orchids (*Orchidaceae*), exhibit another mechanism for concentrating CO_2 at the site of rubisco.
2. CAM is an ancient pathway that likely has been present since the Paleozoic era (570 and 230 Mya) in aquatic species from shallow-water palustrine habitats.
3. The selective factors driving aquatic CAM are autogenic, and CAM is widespread within the plant kingdom across at least 343 genera in 35 plant families comprising ~6% of flowering plant species.
4. The oldest lineage with CAM described to date is represented by Isoetes, a mostly aquatic or semi-aquatic group distributed in oligotrophic lakes or mesotrophic shallow seasonal pools (Keeley 1998).

http://www.mobot.org/mobot/photoessays/guizhou/images/Isoetes_yunguiense.jpg



CO_2 fixation in a CAM plant, pineapple, *Ananas comosus*

A typical well known CAM plant is pineapple

2. CAM Plant Characteristics

1. An important attribute of CAM plants is their capacity to attain high biomass in habitats where precipitation is inadequate, or where evaporation is so great that rainfall is insufficient for crop growth.
2. CAM is generally associated with anatomical features that minimize water loss, such as thick cuticles, low surface-to-volume ratios, large vacuoles, and stomata with small apertures.
3. In addition, tight packing of the mesophyll cells enhances CAM performance by restricting CO_2 loss during the day.
4. Typically, a CAM plant loses 50 to 100 grams of water for every gram of CO_2 gained, compared with 250 to 300 grams for C4 plants and 400 to 500 grams for C3 plants.

3. CO_2 Reduction

1. In CAM plants, the uptake of atmospheric CO_2 takes place at night when stomata are open.
   - At this stage, gaseous CO_2 in the cytosol, coming from both the external atmosphere and mitochondrial respiration, increases levels of HCO_3^- [$CO_2 + H_2O \leftrightarrow HCO_3^- + H^+$].
2. Then cytosolic PEPCase catalyzes a reaction between HCO_3^- and PEP provided by the nocturnal breakdown of chloroplast starch.
3. The resulting four-carbon acid, **oxaloacetate**, is reduced to **malate** which, in turn, proceeds to the acid milieu of the vacuole.
4. During the day, the **malic acid** that was stored in the vacuole at night flows back to the cytosol. Malate decarboxylase (NAD-malic enzyme) acts on malate to release CO_2, which is refixed into carbon skeletons by the Calvin—Benson cycle.

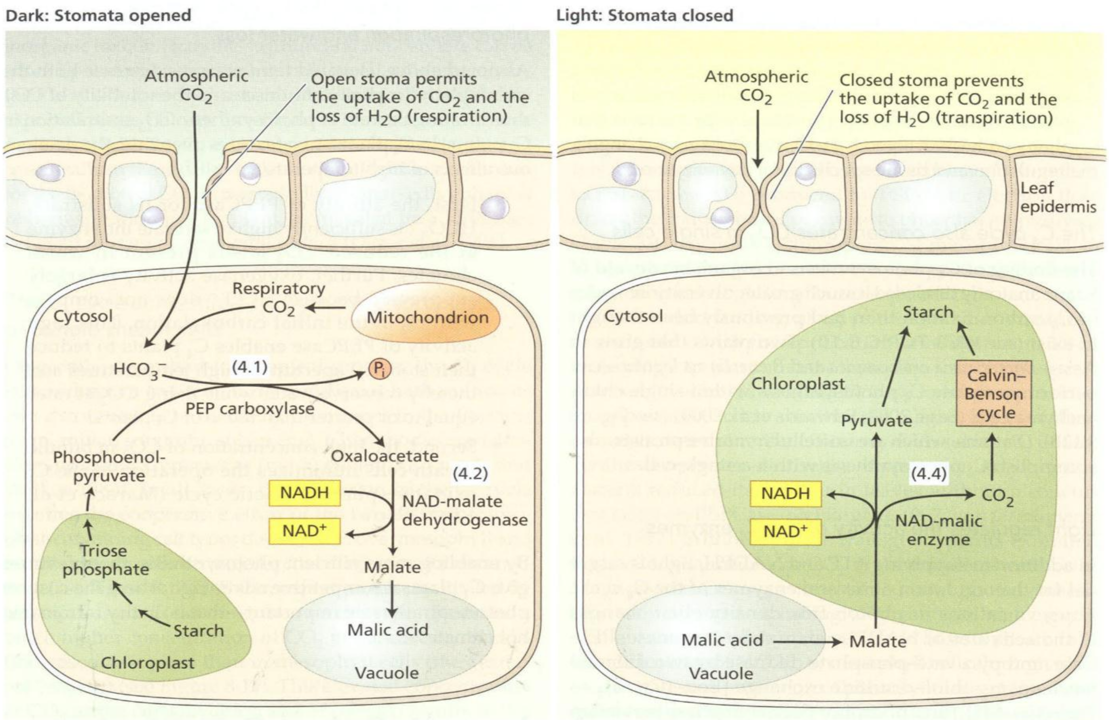


Fig. 8.13 Crassulacean acid metabolism (CAM). In CAM metabolism, CO_2 uptake is separated temporally from fixation via the Calvin—Benson cycle.

REFERENCE

Taiz, L. and Zeiger, E., 2010. Plant Physiology Chapter 8: The Carbon Reaction. Benjamin/Cummings, Company, Inc., Redwood City, California