

## 1. **Pick up** Name Folder

- Pick up name folder and set it up at seat.

## 2. **Sit** with your lab group.

- laptops almost closed (avoid distracting)

## 3. **Clicker** Attendance

- Launch your Top Hat, and get ready to click.

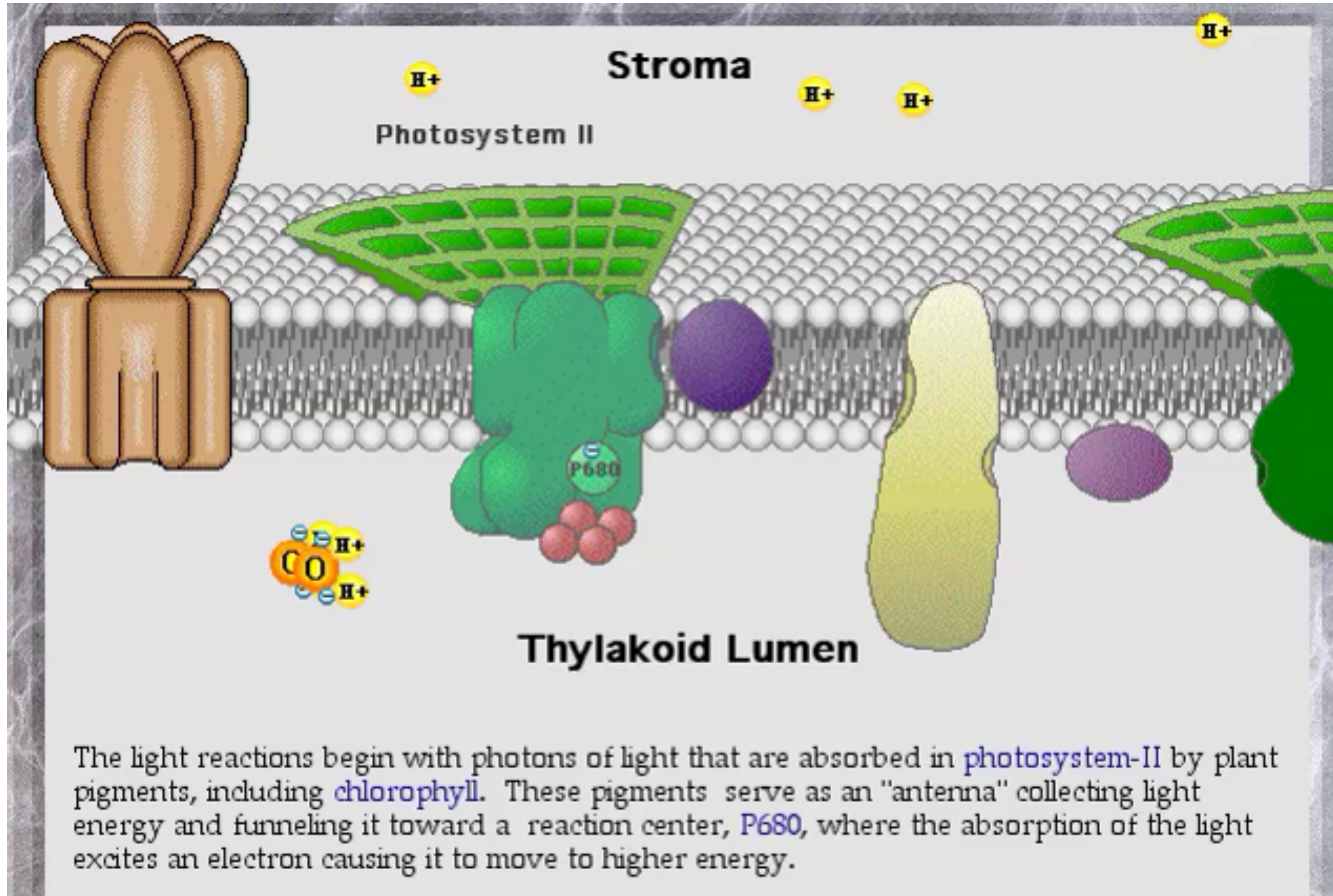
In order for the 8 green photons to be absorbed, they would need to be initially absorbed by carotenoids in order for the energy to be passed down. Two ATP and two NADPH would be made if these 8 photons were absorbed, using the conversions previously used in last week's question. In one run of the calvin cycle, 6 NADPH and 6 ATP are required to generate 1 G3P molecule, so no G3P would be produced from 8 photons, or 2 ATP/2 NADPH.

LAST YEAR'S BEST ANSWER on HOMEWORK

Laptops closed (unless TopHat)



# Biology Theater!



# Announcements

1. **CATME GEA1 feedback** is re-opened, due 11th...
2. **Verbal Final** coverage of topics can start after exam, any attempts that happen before we discuss digestion, get to skip it.
3. **Status Report Talk in week 10 (optional)** replaces Proposal Talk grade IF you wish. Must RSVP email me by Tonight 11pm.
4. **Exam I: Next Monday.** Can bring 3x5 inch card with handwritten notes on both sides. If there's time at end of class today, I can show you possible question formats on exam.

## Week 5

### (Preparing for) Monday's lecture:

**Budgeting homework time (70 min):** Chapter 11 section 11.1 is long with many figures that require thinking and notetaking. The first 2/3's of it is the most important part and just reading that part, which is 3800 words, should take 21 minutes or longer. The data figures are important while those that are just drawings, like 11.3, 11.6, 11.7, 11.9, are not. Of course, when done properly, when you pause to review figures, try Integrating Questions, and take notes, this assignment will take you more like 70 minutes.

**Special Allowance:** Your group can divide up the Trifectas for this lecture.

1. \_\_\_\_\_ **For Monday's lecture**, read **Chapter 11: Photosynthesis**, section 11.1 "Why is paraquat used in America but illegal in Europe?" and as you read it on your computer be sure to take handwritten notes in your lecture notebook. Please read carefully and take good notes for the first 2/3s of the section (**which is about 3800 words**). You can stop taking notes when you come to the yellow box of Integrating Questions #11-14. Then just read the last 1/3 of the section for deeper thought and better understanding.
2. \_\_\_\_\_ **Try to answer some Integrating Questions and Review Questions**. As you read the ICB textbook always attempt to test yourself a little, answer at least one of each set.
3. \_\_\_\_\_ (Trifecta): **Prepare to explain (aloud) Figures 11.1B, 11.2A, B, C, 11.4, 11.5, and 11.8A in class**. \*Special Allowance today\*: If you wish your group can designate who will be responsible for each figure and thus split up the responsibility and reduce the load (Purpose, Methods, Findings).
4. \_\_\_\_\_ **Advanced**: Take a peek at section 8.3, in particular study Figure 1. Take a sneak peek at "Chapter 11: Photosynthesis", section 11.2.

Chapter 11 (ICB)  
Section 1

Photosynthesis (ICB book)

11.1 Why is paraquat used in America but illegal in Europe?

Context: Plants + Animals have many similar enzymes. So herbicides might be targeted to plants only BUT...?

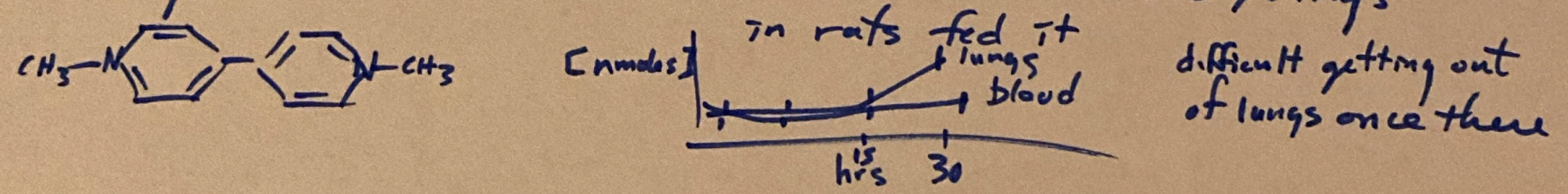
Photosynthesis light rxns are lots like Respiration so herbicide could kill mitochondria too?

L.O.5 -> Describe how plants capture energy from sunlight + convert E.  
• Distinguish three parts of PHS + explain chemical + physical processes

Paraquat

In 2003 EU began using Paraquat herbicide to kill weeds by 2007 EU stopped use. Paraquat poisoning via oral or skin

Figure 11.1 | Paraquat structure + accumulation in blood/lungs



Understanding Photosynthesis

(1774) British Priest Joseph Priestly - testing air candle + plant + sunlight

Purpose: study components in air  
Methods: air tight container, light candle (remotely) -> burns out -> not relight  
Finds: but with sprig of mint + sunlight (few days) -> can relight candle!

(1882) German Theodor Wilhelm Engelmann - testing algae vs rainbow of light x

Purpose: what happened to plants when hit by sunlight?  
Methods: rod-shaped algae (plant) mobile O<sub>2</sub>-loving bacteria (termo) had novel high tech microscope by friend the famous Carl Zeiss!  
Findings: bacteria -> used prism to split light into rainbow moved toward BLUE + RED light areas on algae (absorbance spectrum)

(1937) British Robin Hill (@ Cambridge) | Fig 11.2C

Purpose: ID which part of plant cell does PHS?  
Method: blender vs spinach leaves + centrifuge -> chloroplasts isolated

ICB 11.1 (cont.) PHS

Robin Hill showed isolated chloroplasts could do PHS -> evolve O<sub>2</sub> -> light dosage dependent

- IQs 1-4 | - Paraquat poisoning -> how location of accumulation problematic? -> predict it is water soluble?
1. Priestly trifecta, mint produced what molecule?
  2. Priestly trifecta, mint produced what molecule?
  3. Engelmann trifecta
  4. Hill trifecta "Hill reaction" (Colin's boxes + lights)

Paraquats charge important - likely substituting for PQ

Fig 11.3 | - Anatomy of Chloroplast + PS

IQs 5-9 | 1. colors chl absorbs, eyes see it as? why plants look green, if have yellow pigments  
Note: thylakoids can be stacked or unstacked dynamically

(1954) American Daniel Arnon (UC Berkeley)

Figure 11.4 | Purpose: measuring the rate of "splitting H<sub>2</sub>O", making ATP, fix CO<sub>2</sub>  
Methods: use an inhibitor at different concentrations  
- exposed isolated chloroplasts  
- detected amount O<sub>2</sub> produced  
Findings: H<sub>2</sub>O splitting not effected BUT ATP + CO<sub>2</sub> fix were impacted but different  
-> suggested all three separate dose-response curves

Q: If reviewing Arnon's paper, ask for additional experiments? dose-response


(1973) German Hans Heldt -> ATP production

Figure 11.5 | Purpose: study mechanism behind ATP production at thylakoid mem  
Methods: Track pH inside + outside with pointy micro electrodes (lumen/space) (stroma) simultaneously of a single chloroplast  
- Turn off/on light / dark  
Findings: light ON -> stroma loses H<sup>+</sup> + thylakoid gains them (pH↓), repeated (pH↑)

then Robin Hill -> photobleaching supported two photosystems P680 P700  
Reading-reviews Z-scheme path + carriers of light rxns + cyclic sch + talks about movement / pumping of protons at cytochrome

Figure 11.8 | ATP synthase "rotational catalysis". chloroplasts mitochondria + prokary ATP synthase

# Understanding Photosynthesis

1774 British Priest Joseph Priestly - testing air  mint candle + plant + sunlight

Purpose: study components in air

Methods: air tight container, light candle (remotely) → <sup>few minutes</sup> burns out → not relight

Findings: but with sprig of mint + sunlight (few days) → can relight candle!

1882 German Theodor Wilhelm Engelmann - testing algae vs rainbow of light x

Purpose: what happened to plants when hit by sunlight?

Methods: rod-shaped algae (plant) mobile  $O_2$ -loving bacteria (termo)

had novel high tech microscope by friend the famous Carl Zeiss!

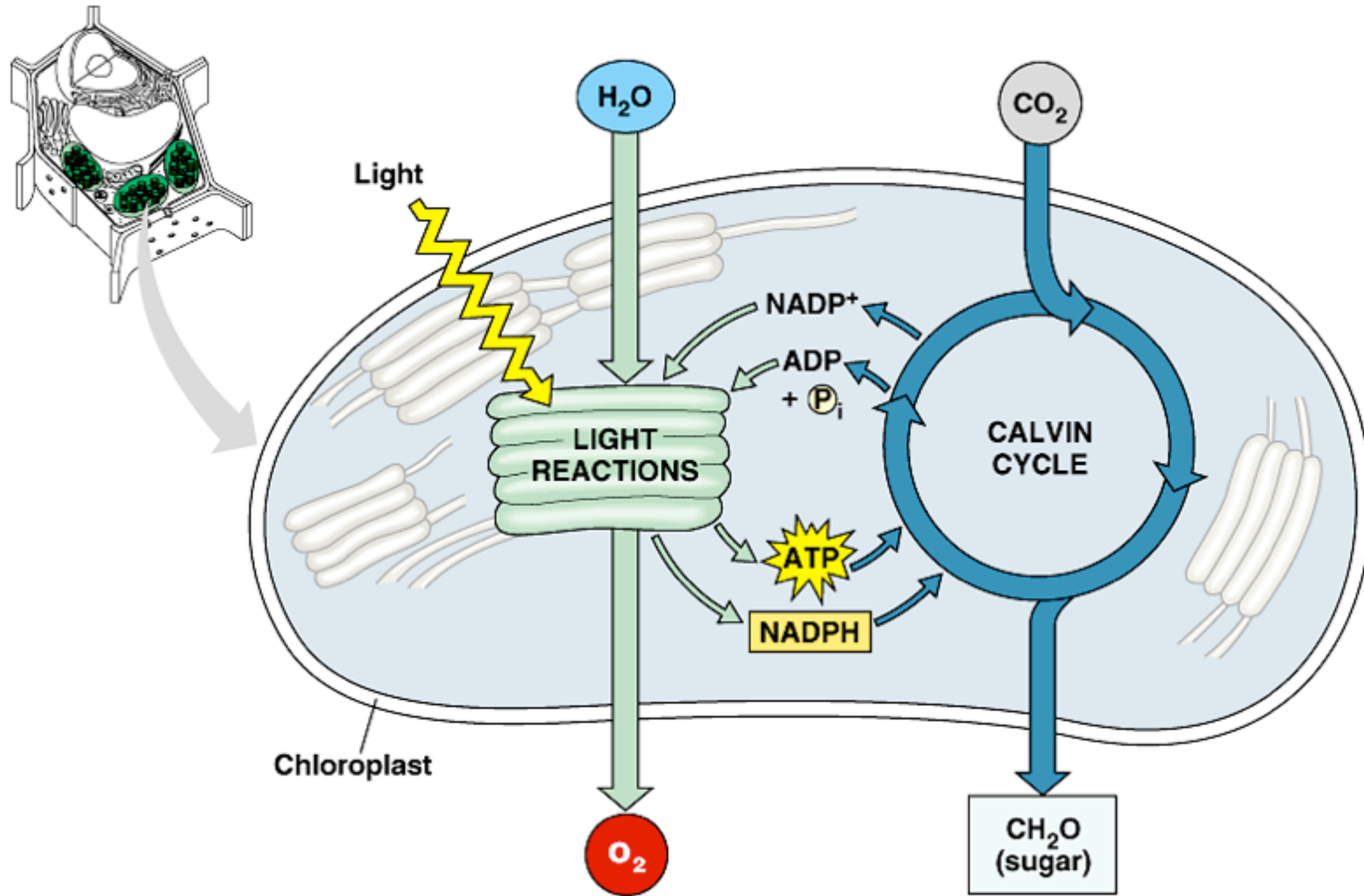
Findings: bacteria → used prism to split light into rainbow <sup>living!</sup> moved toward BLUE + RED light areas on algae (absorbance spectrum)

1937 British Robin Hill (@ Cambridge) Fig 11.2C

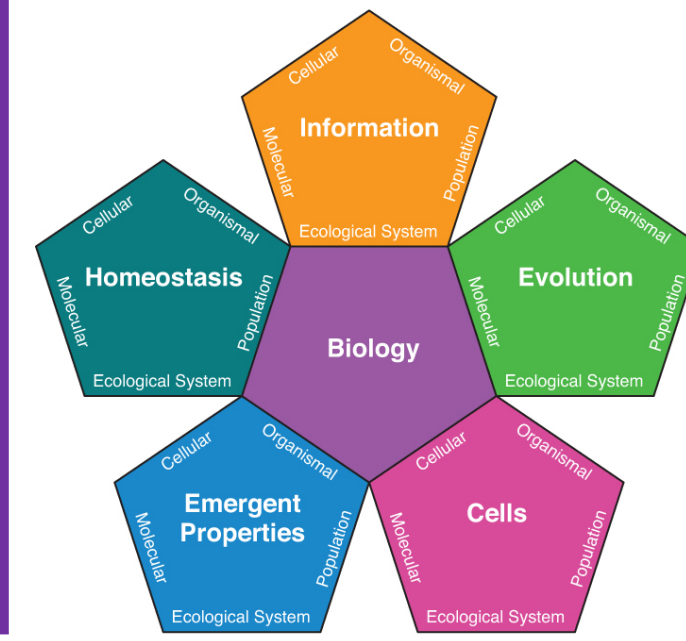
Purpose: ID which part of plant cell does PHS?

Method: blender vs spinach leaves + centrifuge → chloroplasts isolated

Back to Photosynthesis...



# *Integrating Concepts in Biology*



## Chapter 11: **Photosynthesis**

11.1 Why is paraquat used in America  
but illegal in Europe?

by A. Malcolm Campbell, Laurie J. Heyer, &  
Christopher Paradise

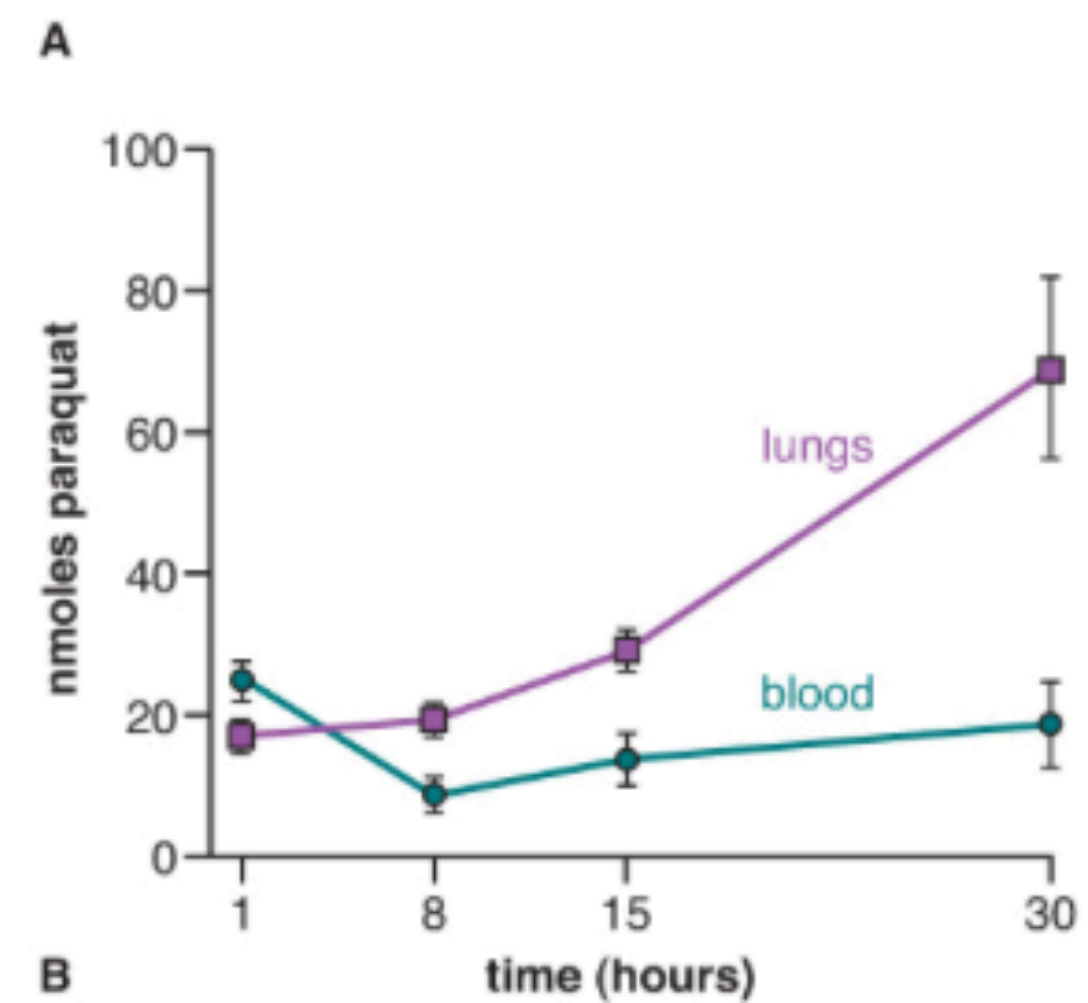
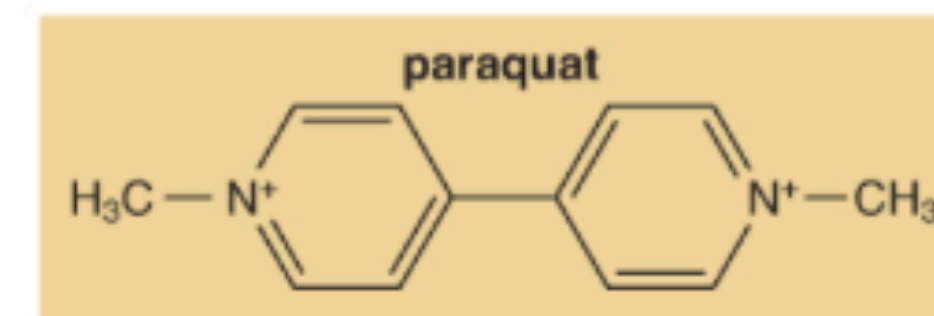
## 11.1 Why is paraquat used in America but illegal in Europe?

- Context: Plants and animals contain many similar enzymes. Herbicides are designed to kill only plants, but does our common eukaryotic ancestry put your health at risk too?
- Major themes: Biological systems use feedback mechanisms to regulate and maintain optimal conditions; time-dependent processes regulate biological systems; life requires organization, which is energy dependent; and a biological system's size and environment influence how it addresses physical and chemical challenges.
- Bottom line: Plant molecules that harvest light energy to make new chemical bonds are very similar to molecules in cellular respiration, which means that herbicides might be toxic to animals.

### Biology Learning Objectives

- Describe how plants capture energy from sunlight and convert that energy into new forms of potential energy.
- Distinguish the three parts of photosynthesis, and explain the overall chemical and physical processes involved in each one.

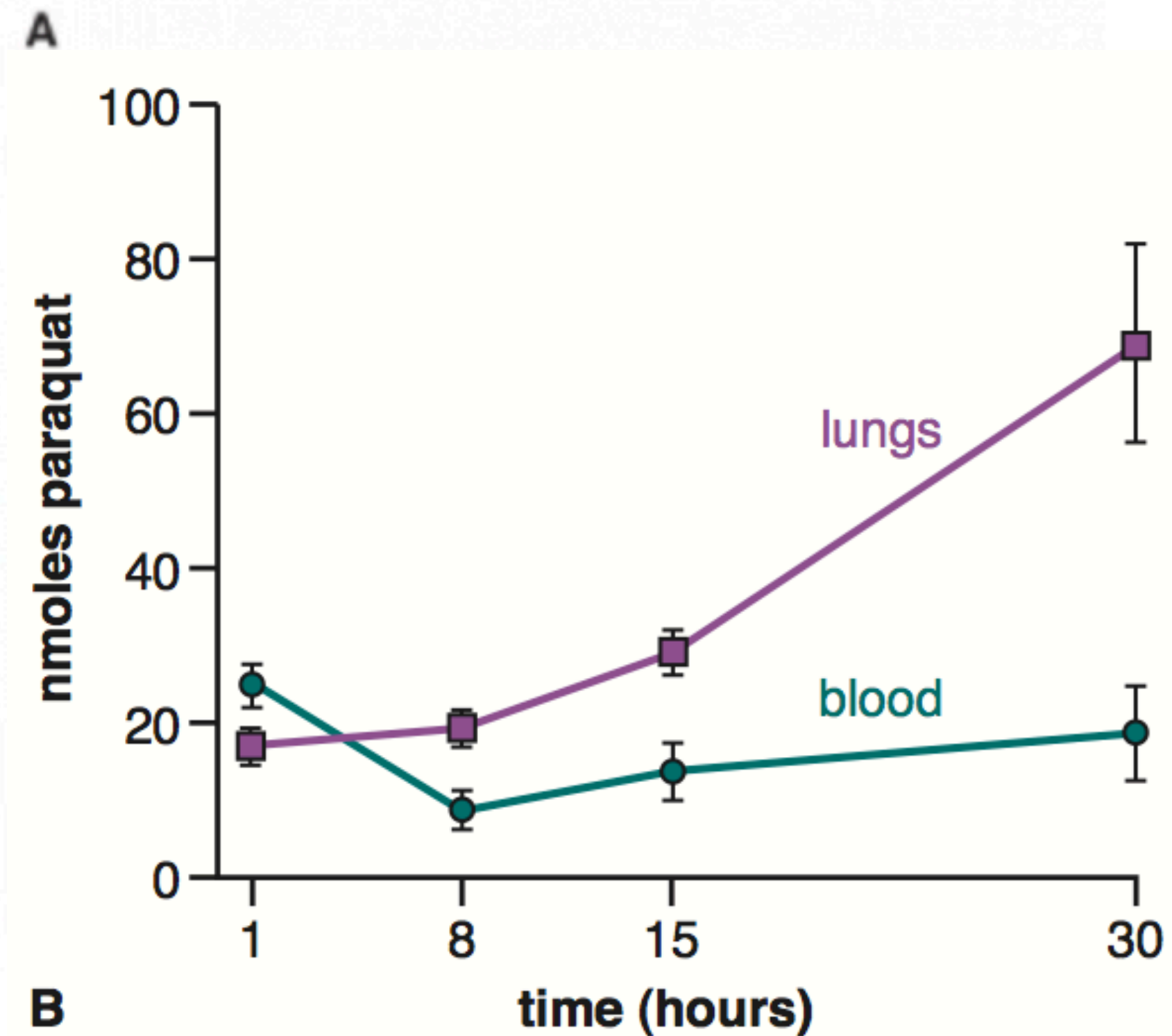
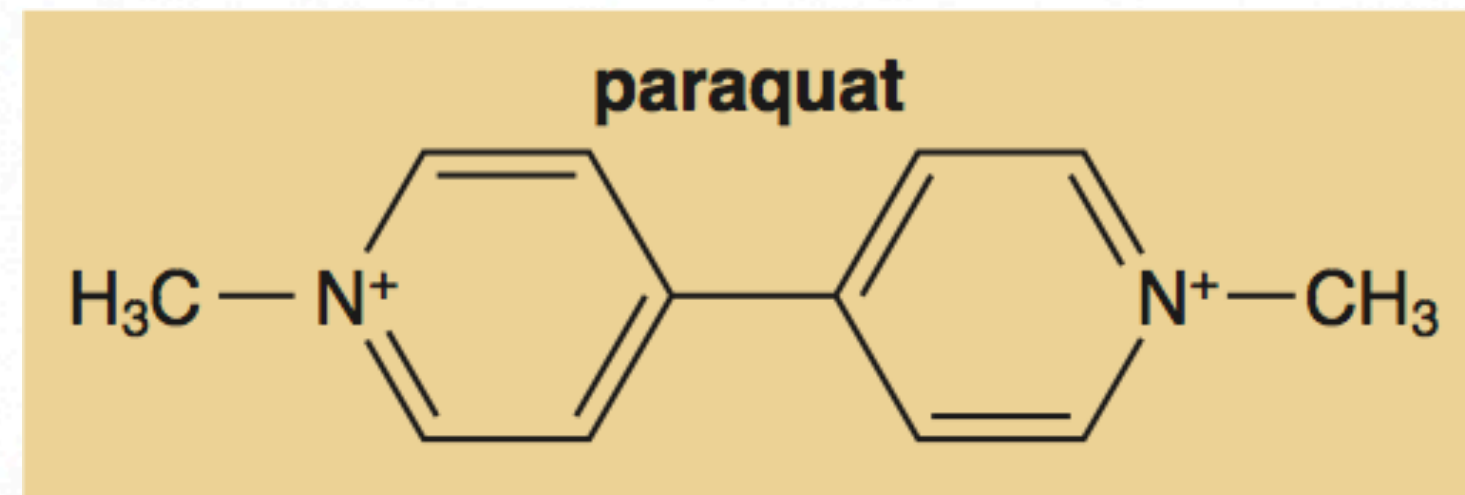
In 2003, farmers in the European Union (EU) began using the most common herbicide on the planet—paraquat (Figure 11.1A). Paraquat is used in over 100 countries to kill unwanted plants, especially weeds that reduce crop yields. In 2007, an EU Court blocked the use of paraquat in Europe. Farmers in the EU are not allowed to use paraquat, because it “[f]ails to satisfy the requirement of protection of human health.” Most cases of paraquat poisoning are due to oral ingestion, but some people have died due to absorption through the skin. How does paraquat work on plants? Can an herbicide kill plants and humans? Pathologists quantified the level of paraquat in lab rats that had consumed potentially lethal doses of the herbicide (Figure 11.1B). Physicians can try to lower the harm of ingested paraquat, but lowering the blood level of paraquat is easier than stopping the accumulation in lungs. To understand how paraquat works in plants and how this might affect humans, you need to understand photosynthesis first.



## **Biology Learning Objectives**

- Describe how plants capture energy from sunlight and convert that energy into new forms of potential energy.
- Distinguish three independent parts of photosynthesis, and explain the overall chemical and physical processes involved in each one.

# Trifecta



**Figure 11.1** Paraquat and its mammalian consequences. **A**, Molecular structure of the herbicide paraquat. **B**, Paraquat levels in the blood (per mL) and lungs (per gram) of rats fed paraquat. Error bars are standard error;  $n = 5$  rats per time point. Panel A from public domain, common knowledge. Panel B modified from LL Smith, 1985; his figure 1. Smith, L. L. 1985. Paraquat Toxicity. Philosophical Transaction of the Royal Society of London. Vol. B 311: 647 – 657. Reprinted with permission from The Royal Society. Copyright © 1985.



# Trifecta

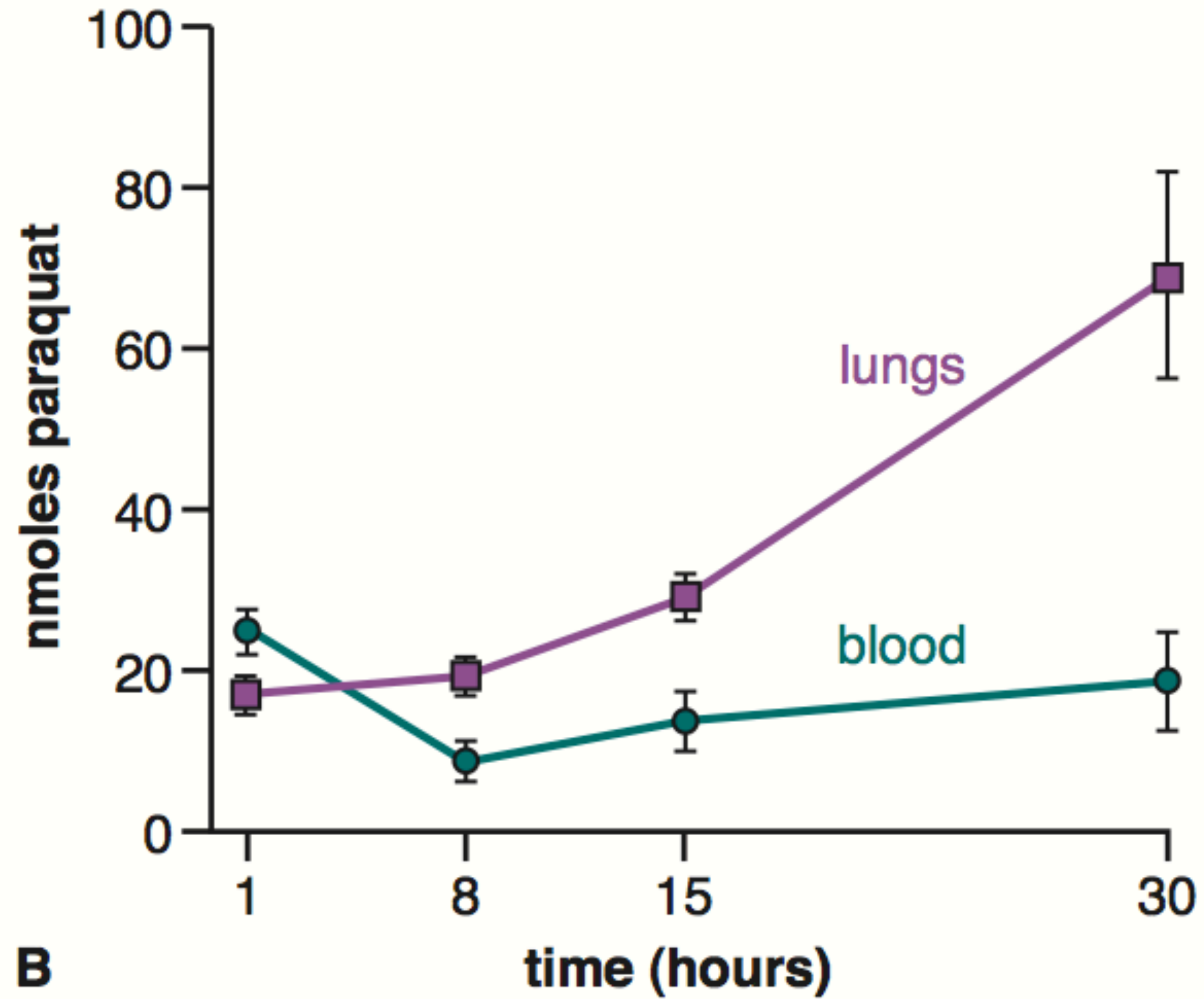


Fig. 11.1

# Paraquat Affects Mammals

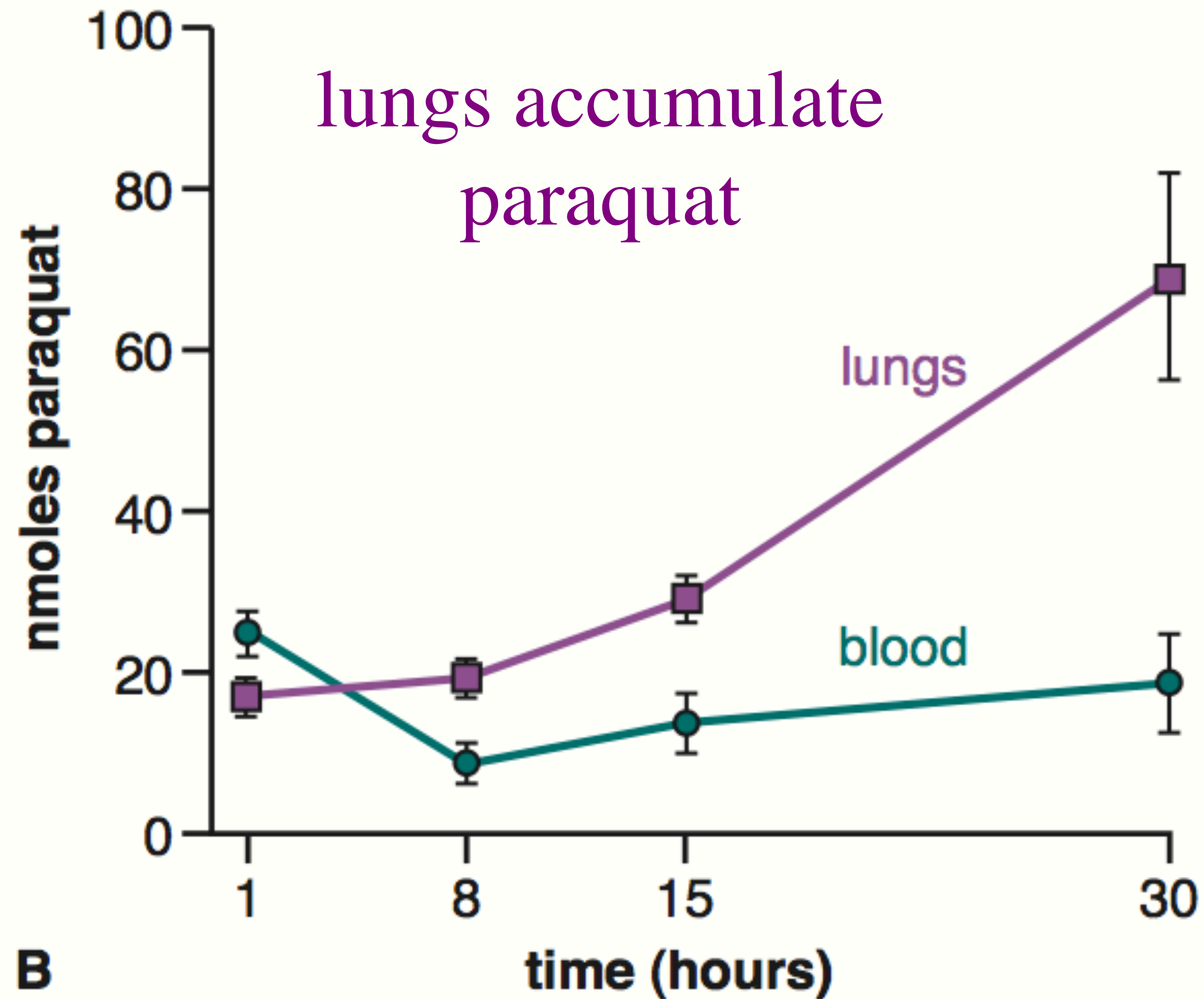


Fig. 11.1

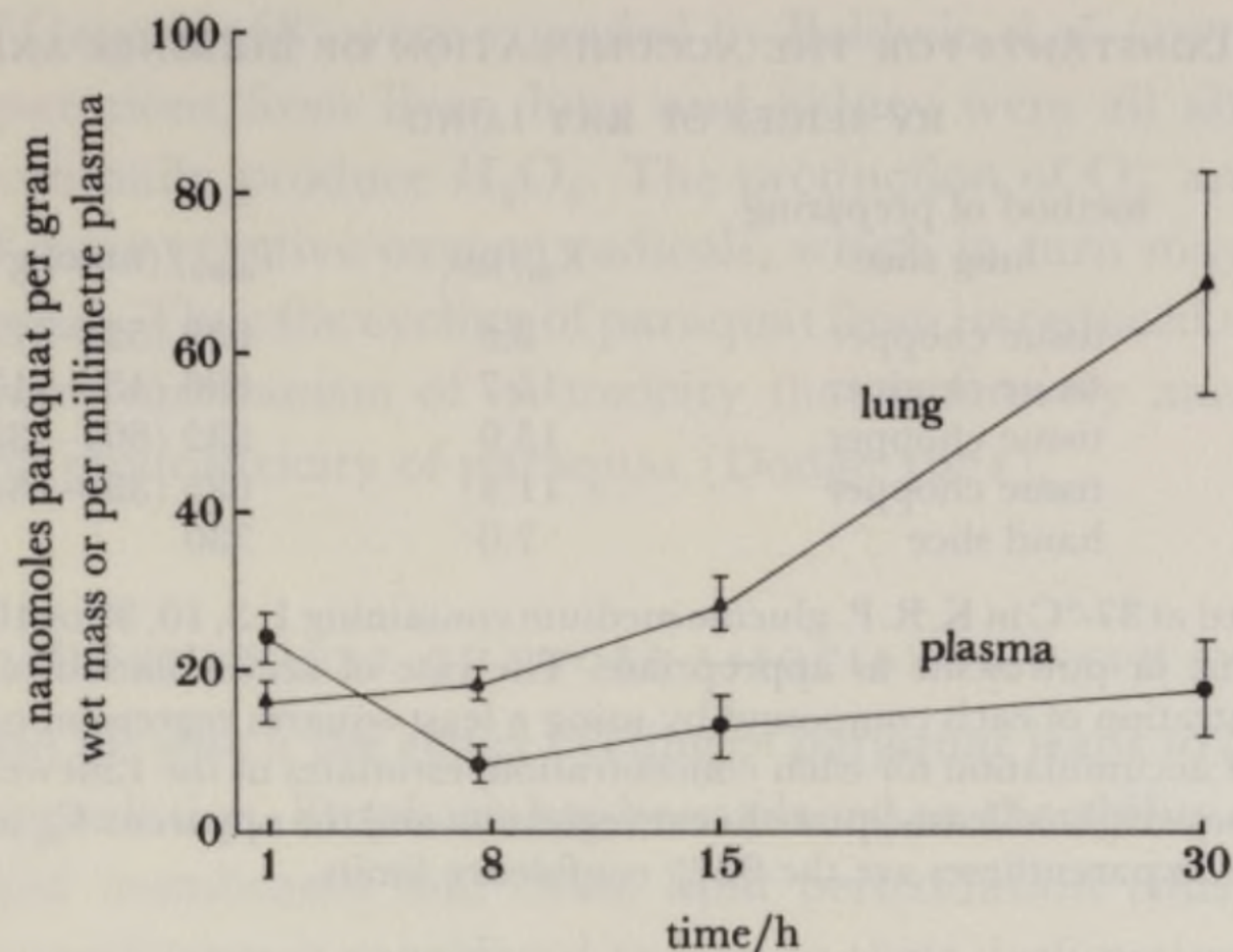
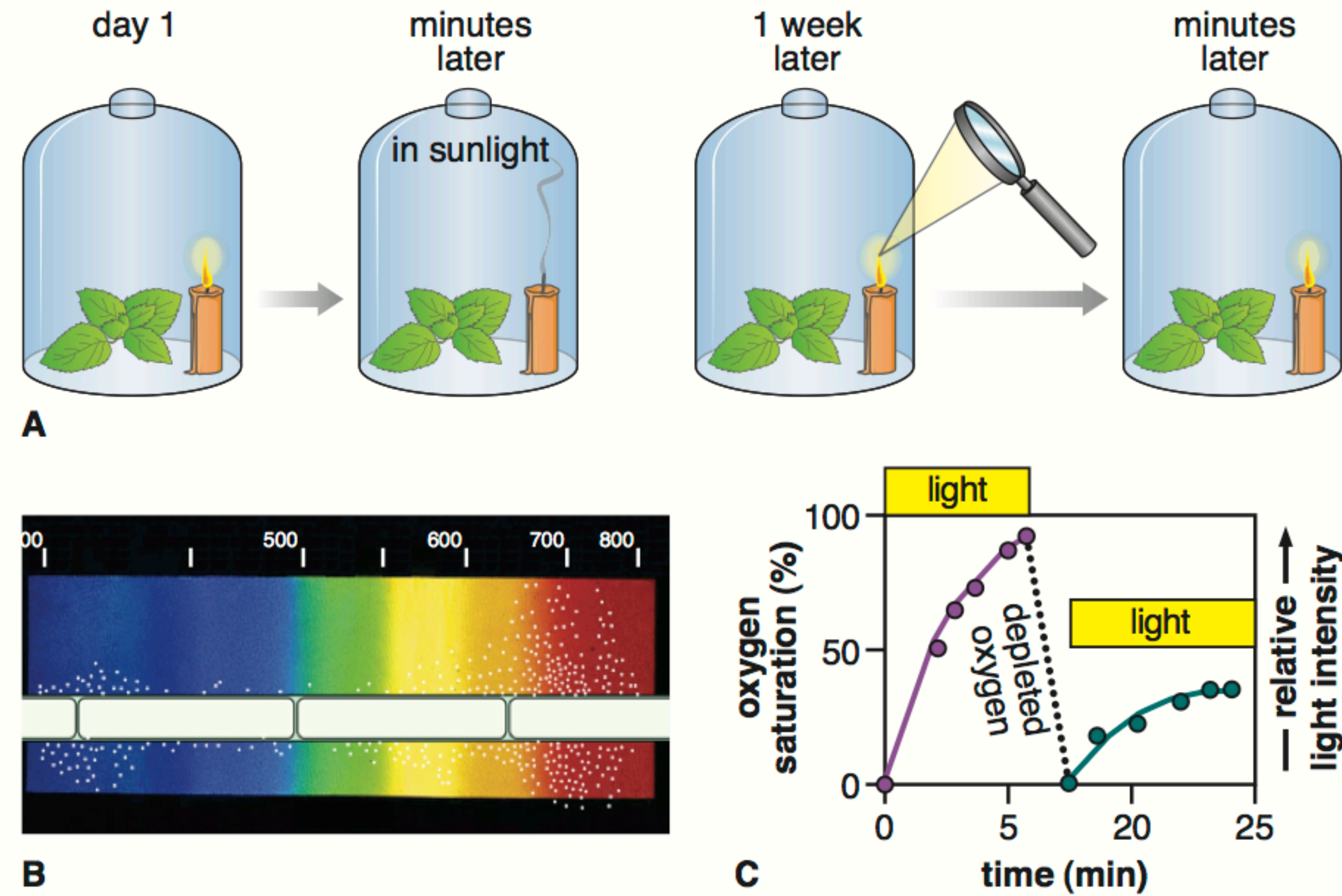


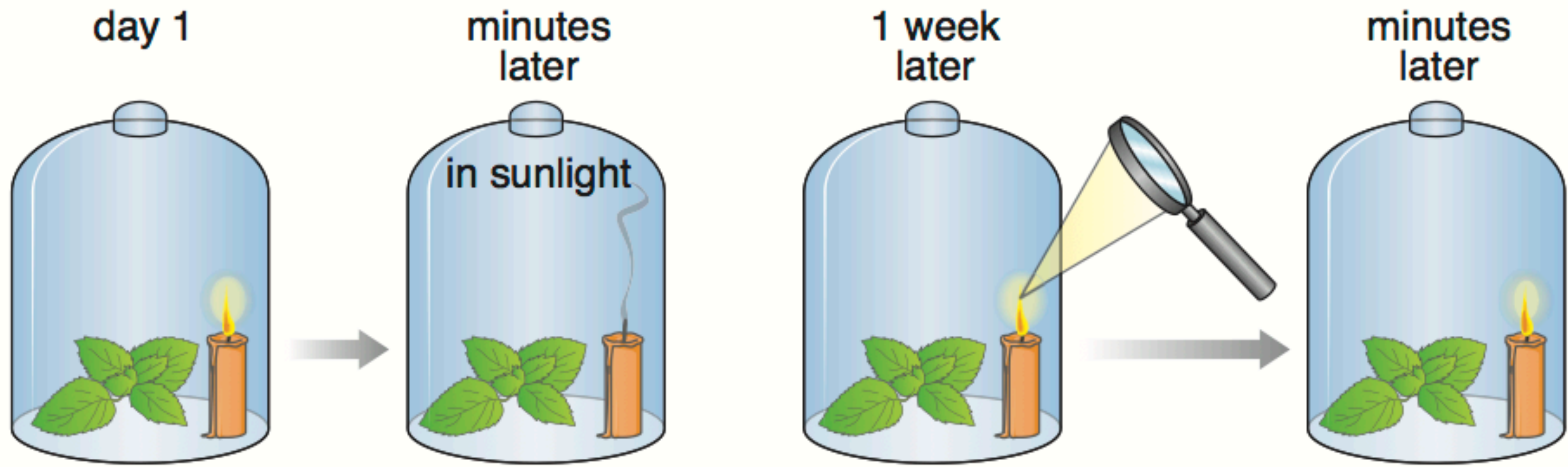
FIGURE 1. The level of paraquat in the lung and plasma of rats given  $680 \mu\text{mol}$  of paraquat per kilogram of body mass orally. Points on the graph represent the mean  $\pm$  s.e. (standard error) with at least five rats per time point. Rate of uptake: 4–6 nmol per gram wet mass per hour.

with the exception of brain slices, which could accumulate paraquat, but to a much lesser extent than lung slices (Rose *et al.* 1976).

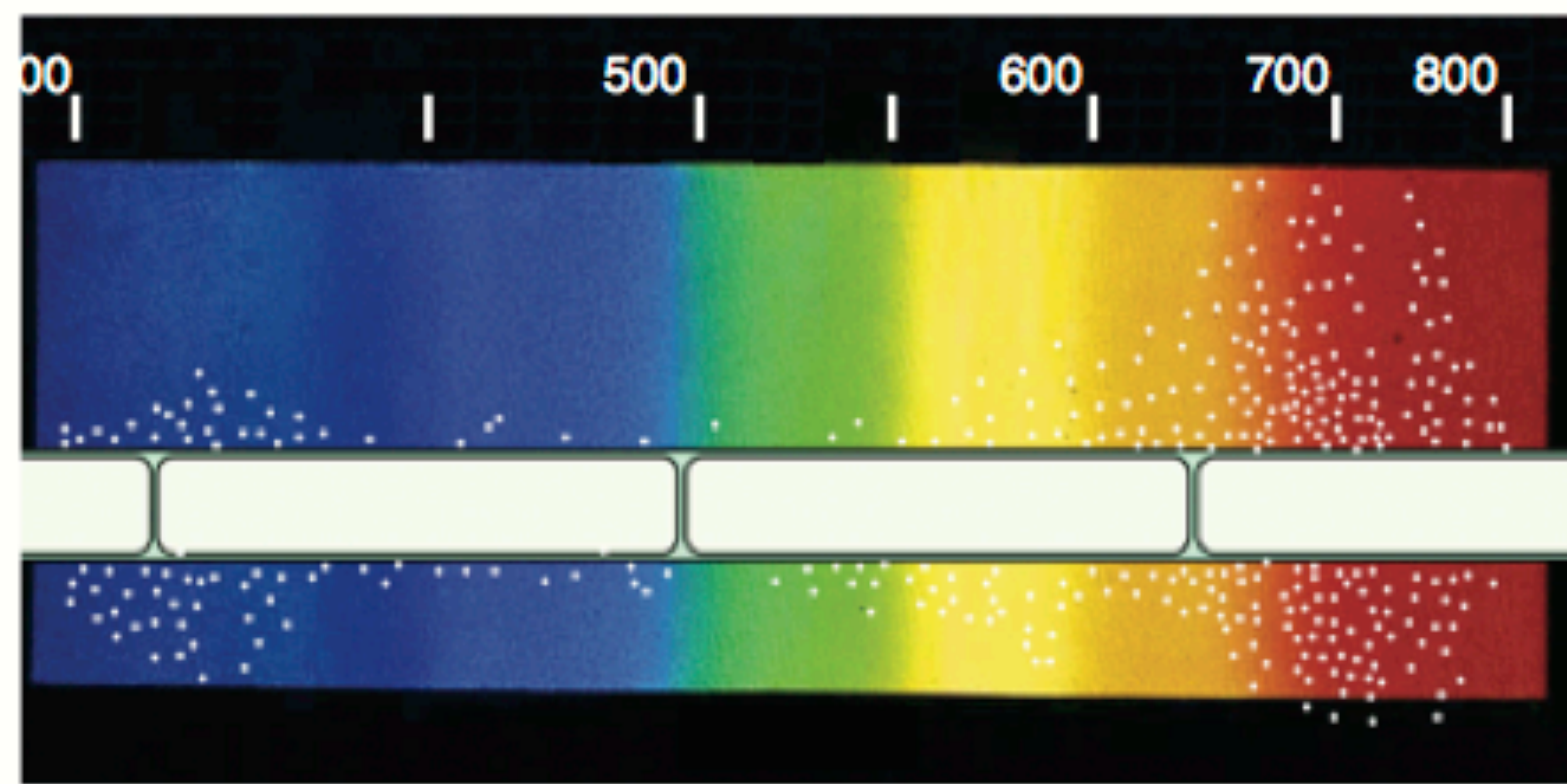
# Trifecta



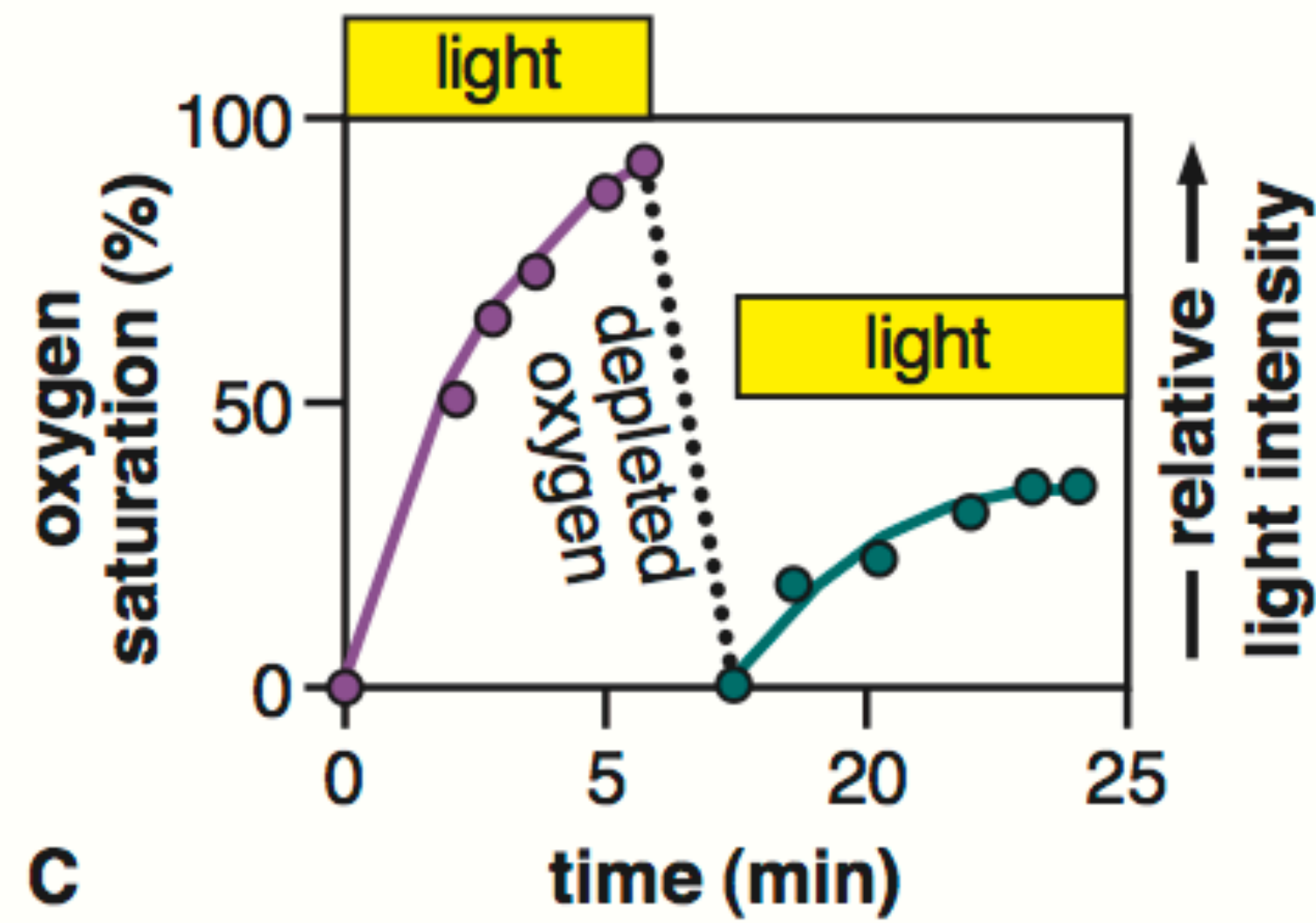
**Figure 11.2** Plants produce oxygen. **A**, Priestley's 1774 experiment showing that plants in the sunlight can replenish depleted air. **B**, Engelmann's 1882 experiment showed oxygen-loving bacteria (white spots) moved to the portion of algae cells (horizontal rectangles) exposed to particular wavelengths of light. The original drawing was in black and white; color has been added for clarity. **C**, Hill's experiment measured oxygen production (y-axis, left side) from purified chloroplasts exposed to different intensities of light (yellow boxes, y-axis right side). Panel A original art based on Joseph Priestley's 1774 written description. Panel B modified from Engelmann, 1882, his figure 1. Kamen, Martin D. 1986. On creativity of eye and ear: a commentary on



**A**



**B**



**C**

Fig. 11.2

A: from Priestly, 1774; B: from Engelmann, 1882; C: modified from Hill, 1937  
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# Trifecta

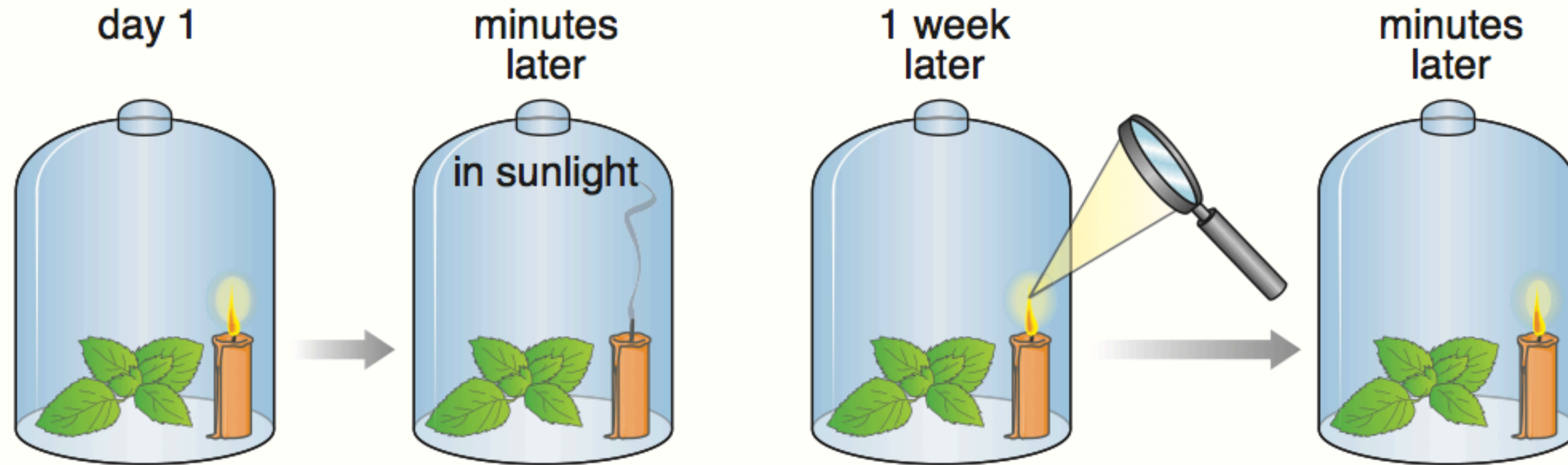
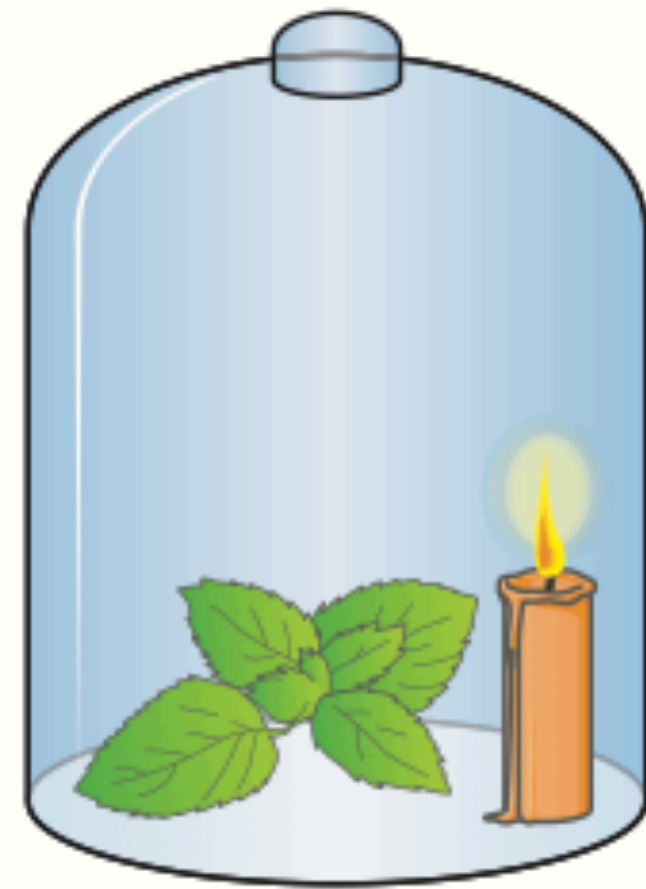


Fig. 11.2

A: from Priestly, 1774; B: from Engelmann, 1882; C: modified from Hill, 1937  
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# Experiments Determine Oxygen Source

day 1



light flame  
under glass  
with plant

Fig. 11.2

# Experiments Determine Oxygen Source

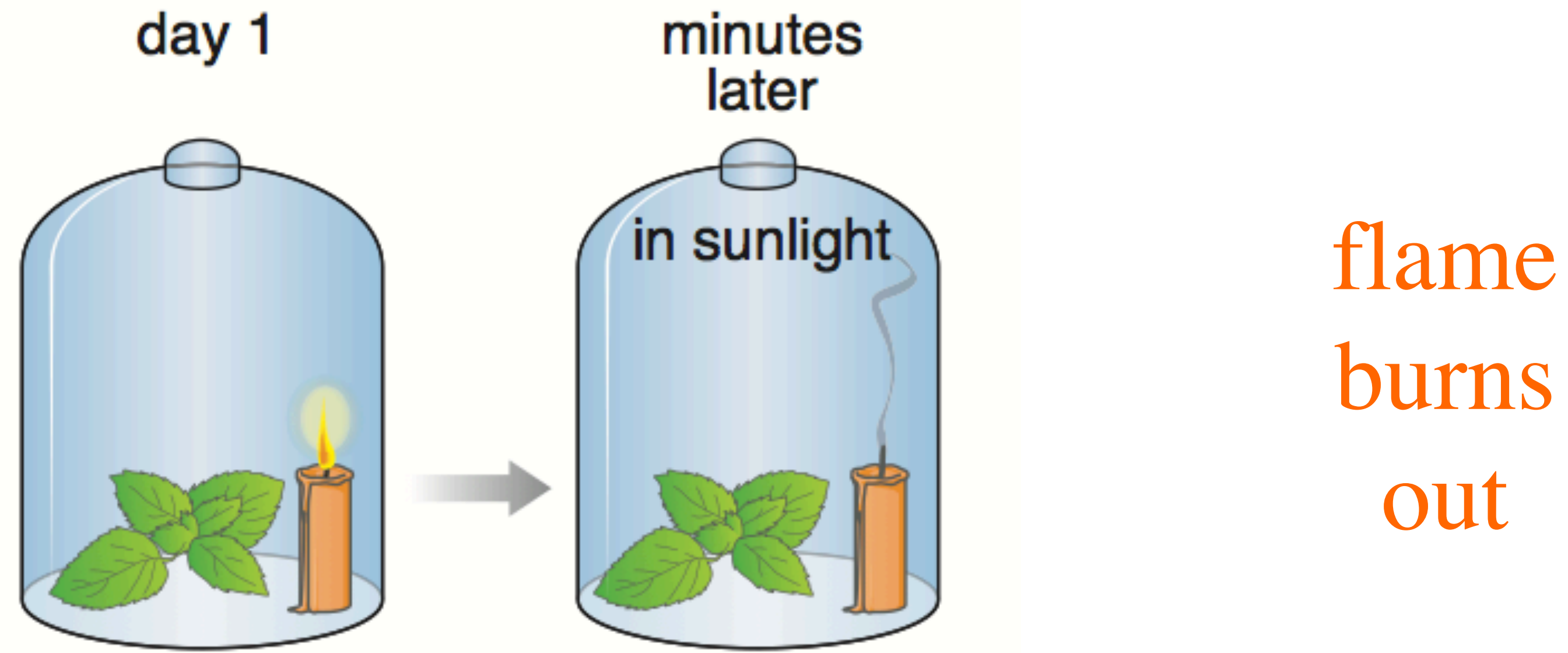


Fig. 11.2

# Experiments Determine Oxygen Source

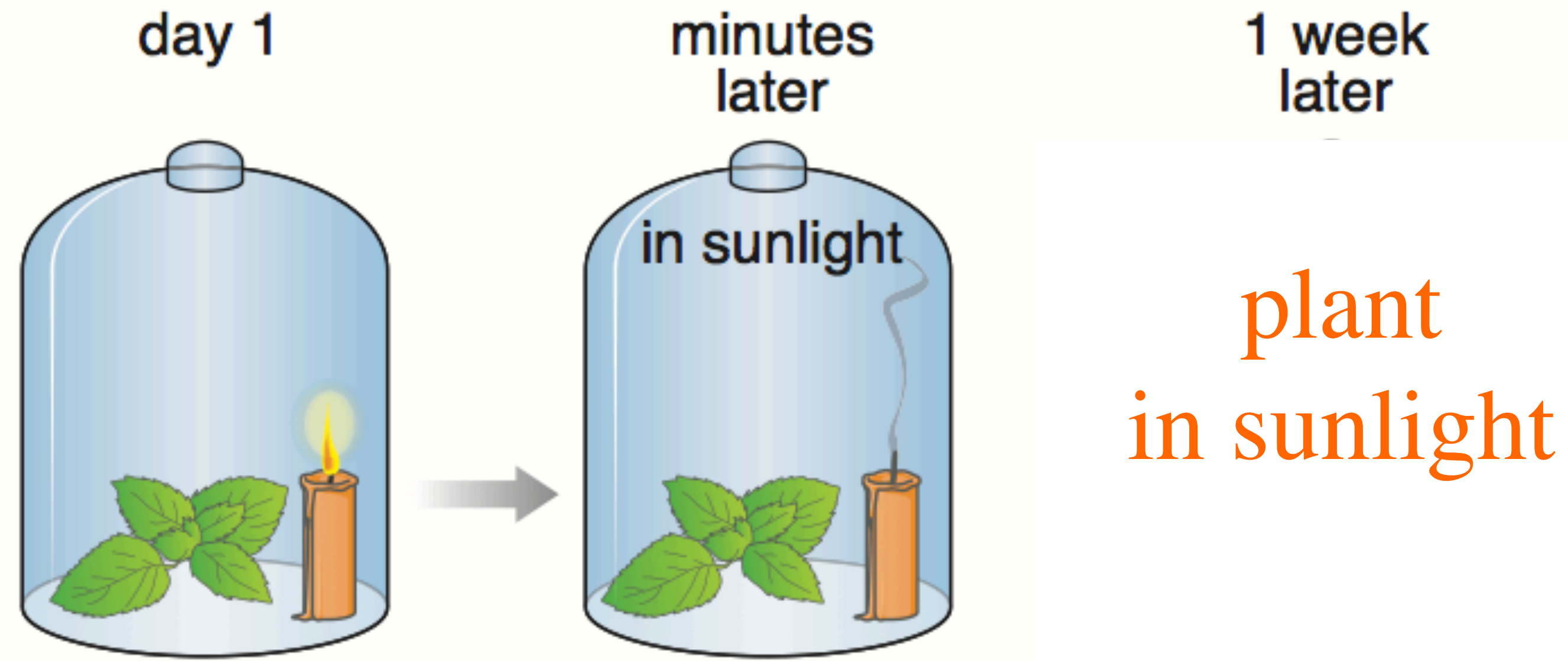
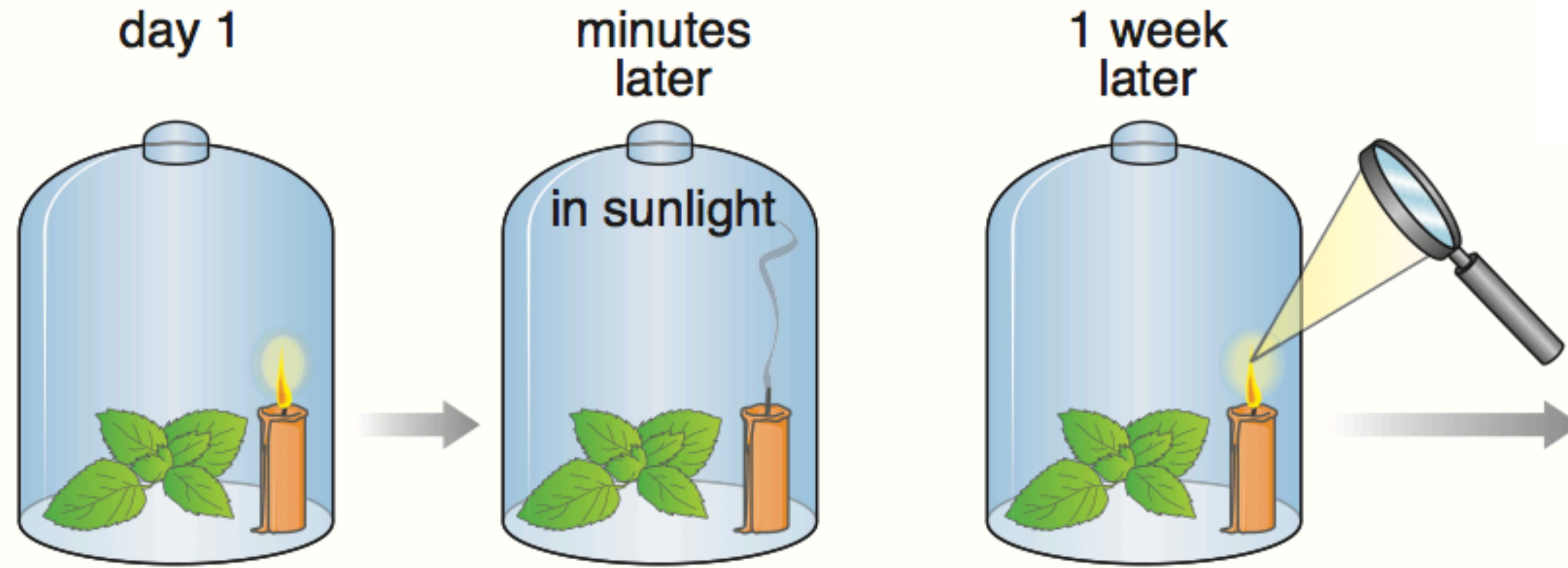


Fig. 11.2

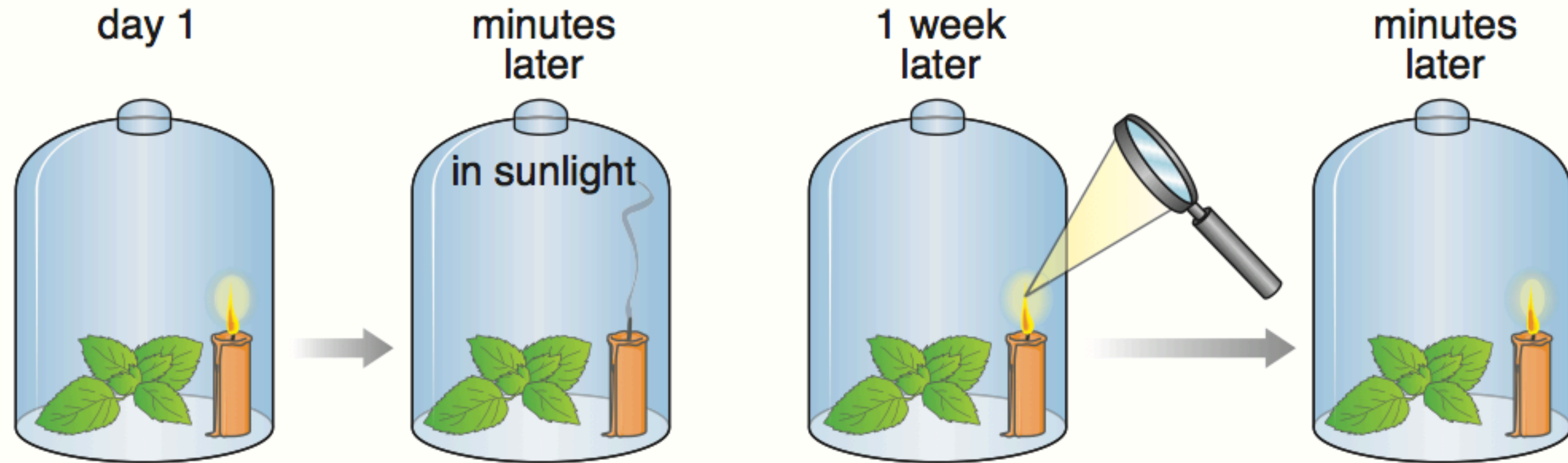
# Experiments Determine Oxygen Source



intensify  
sunlight  
on wick

Fig. 11.2

# Experiments Determine Oxygen Source



flame  
burns  
again

Fig. 11.2

# Trifecta

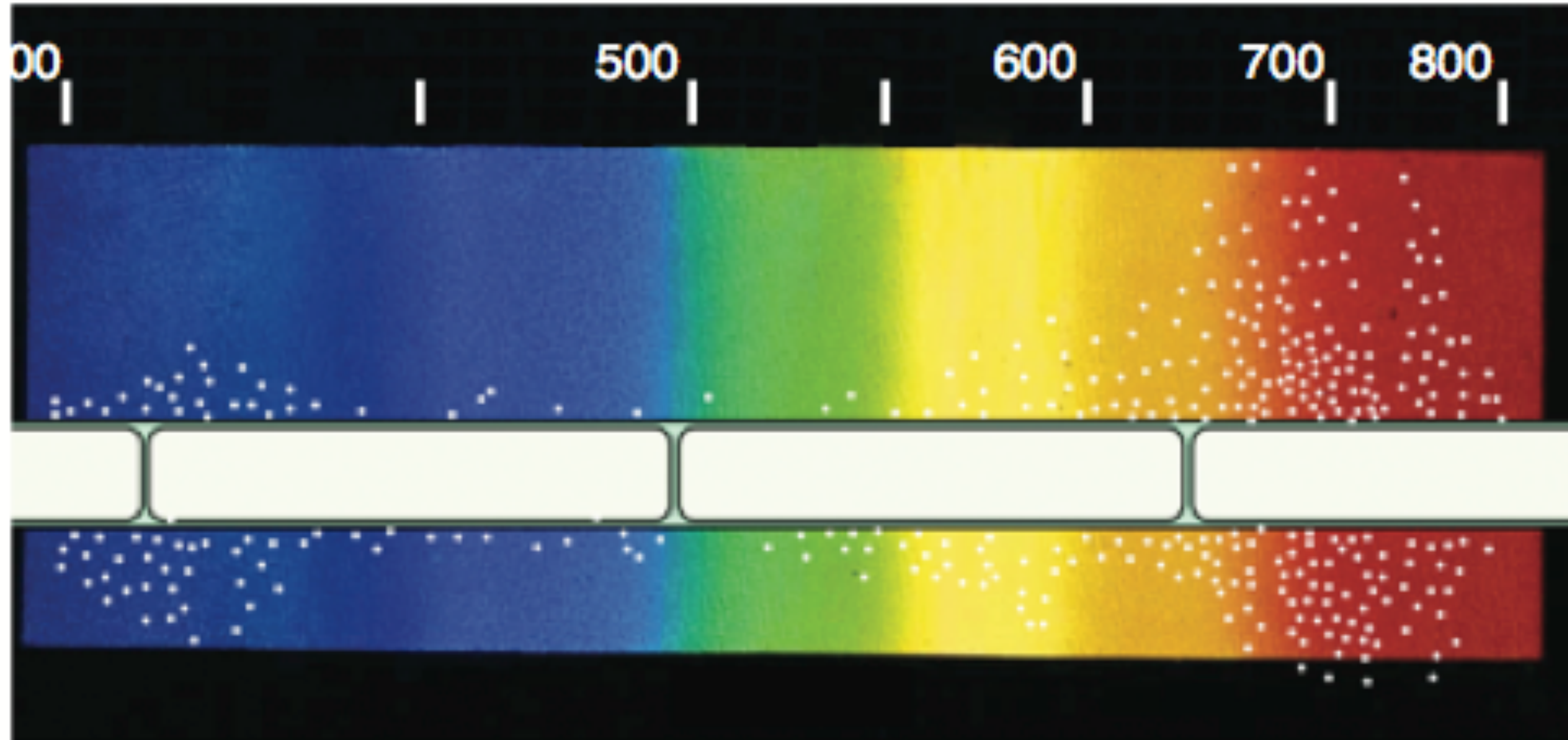


Fig. 11.2

A: from Priestly, 1774; B: from Engelmann, 1882; C: modified from Hill, 1937  
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# Biological Detection of Oxygen Source

green alga viewed through microscope

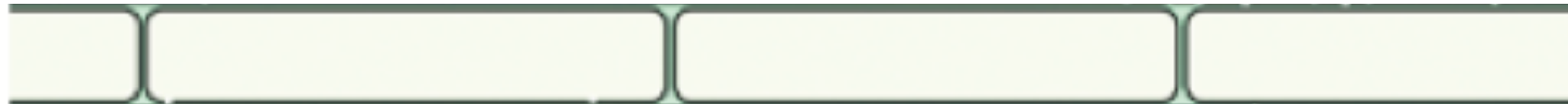


Fig. 11.2

# Biological Detection of Oxygen Source

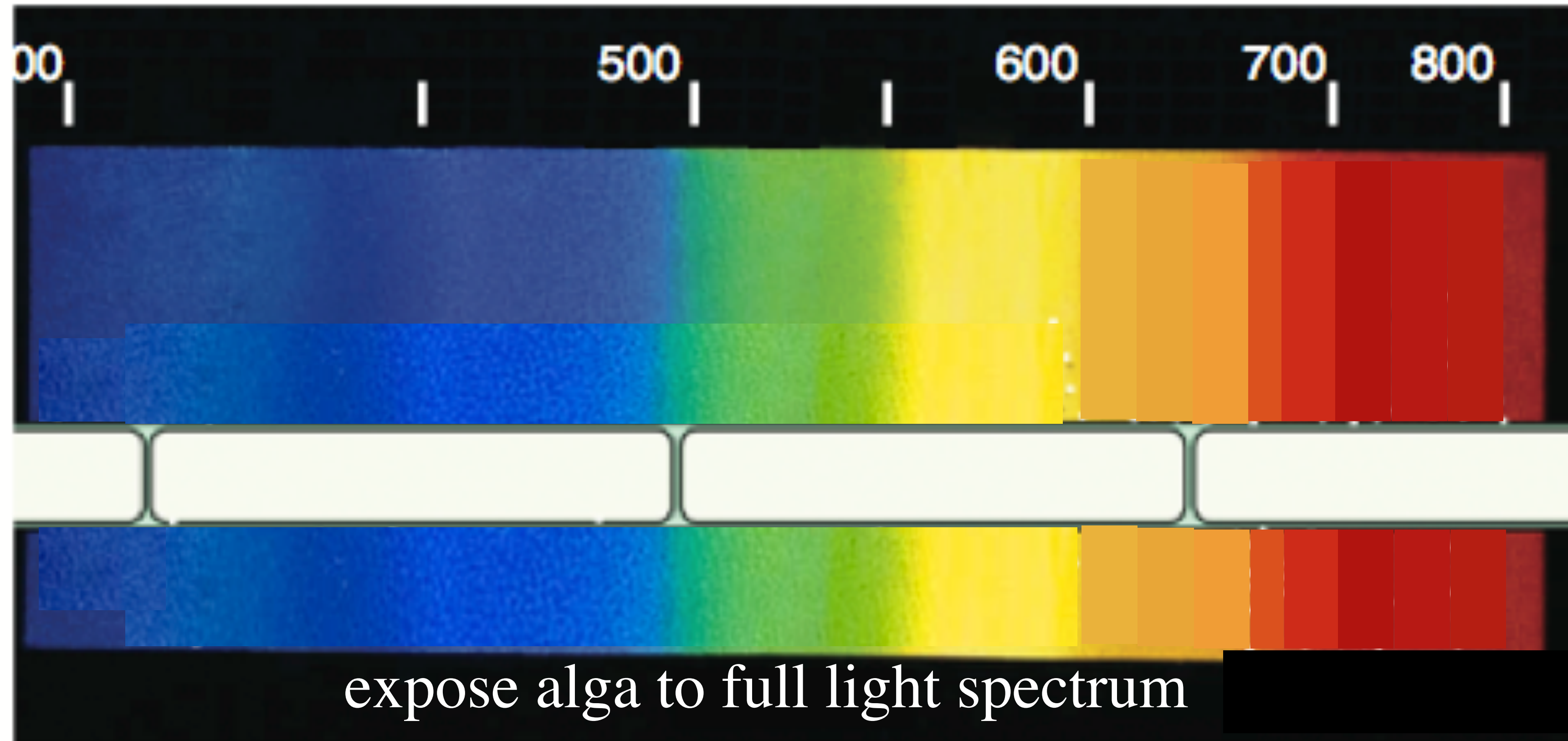


Fig. 11.2

A: from Priestly, 1774; B: from Engelmann, 1882; C: modified from Hill, 1937  
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# Biological Detection of Oxygen Source

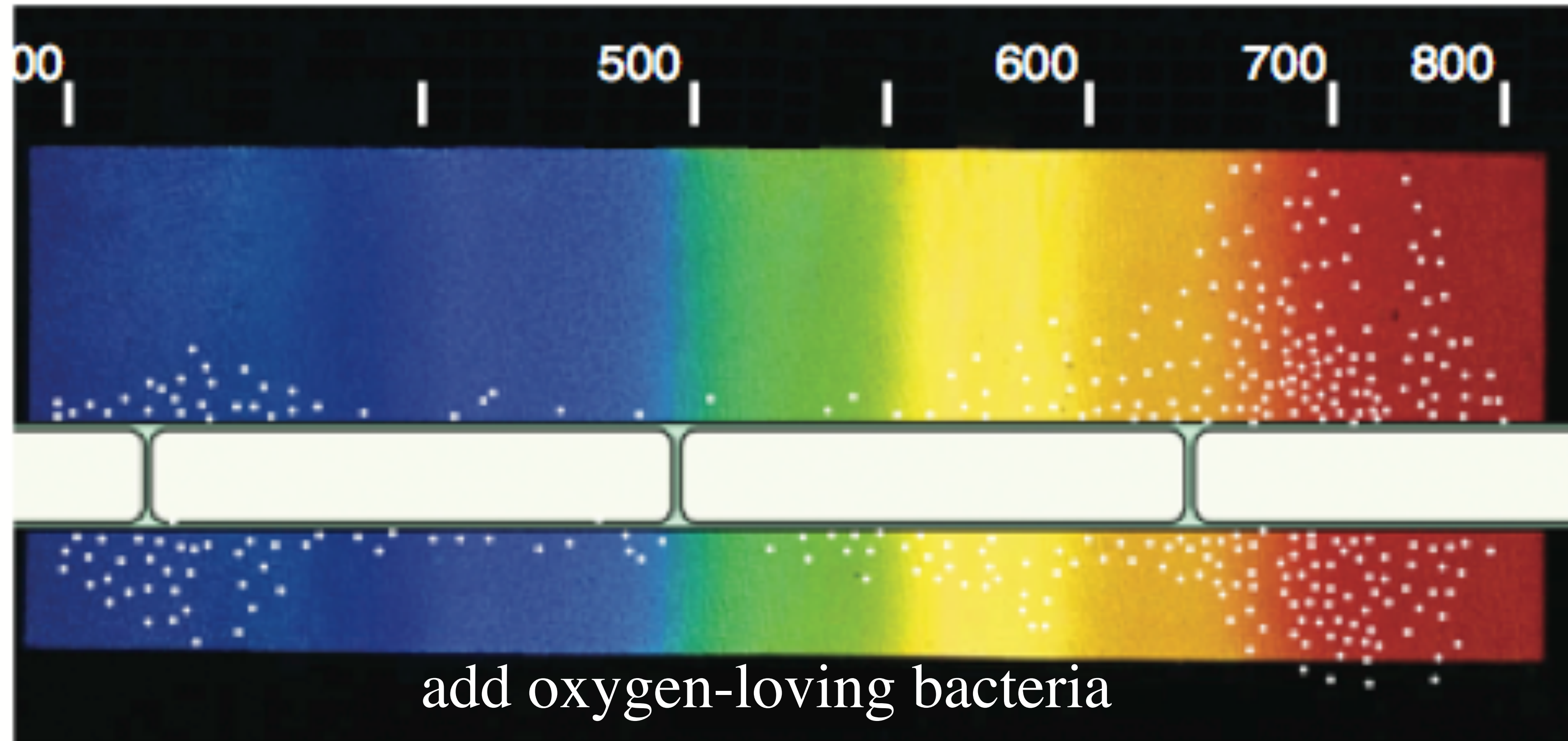


Fig. 11.2

A: from Priestly, 1774; B: from Engelmann, 1882; C: modified from Hill, 1937  
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# Biological Detection of Oxygen Source

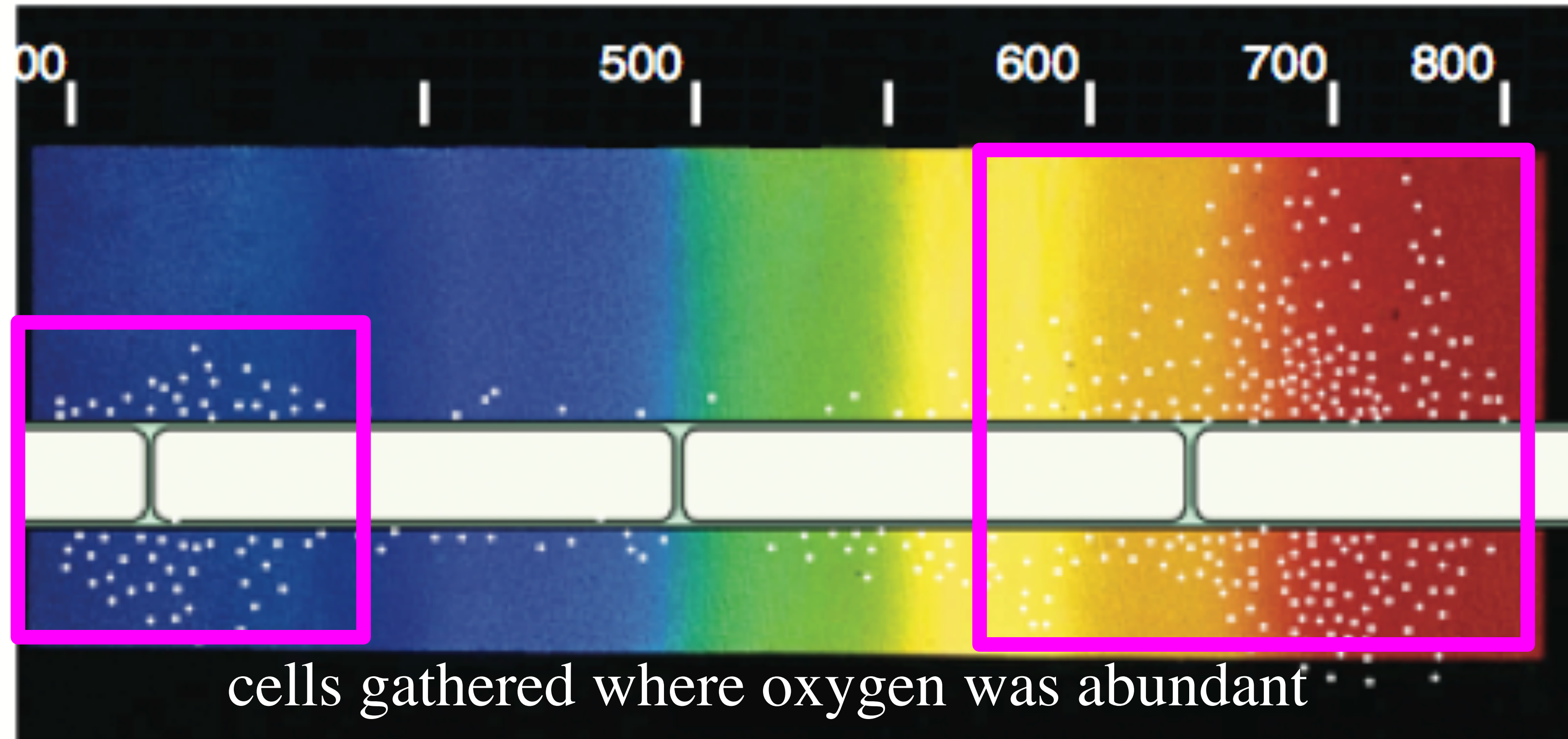


Fig. 11.2

# Trifecta

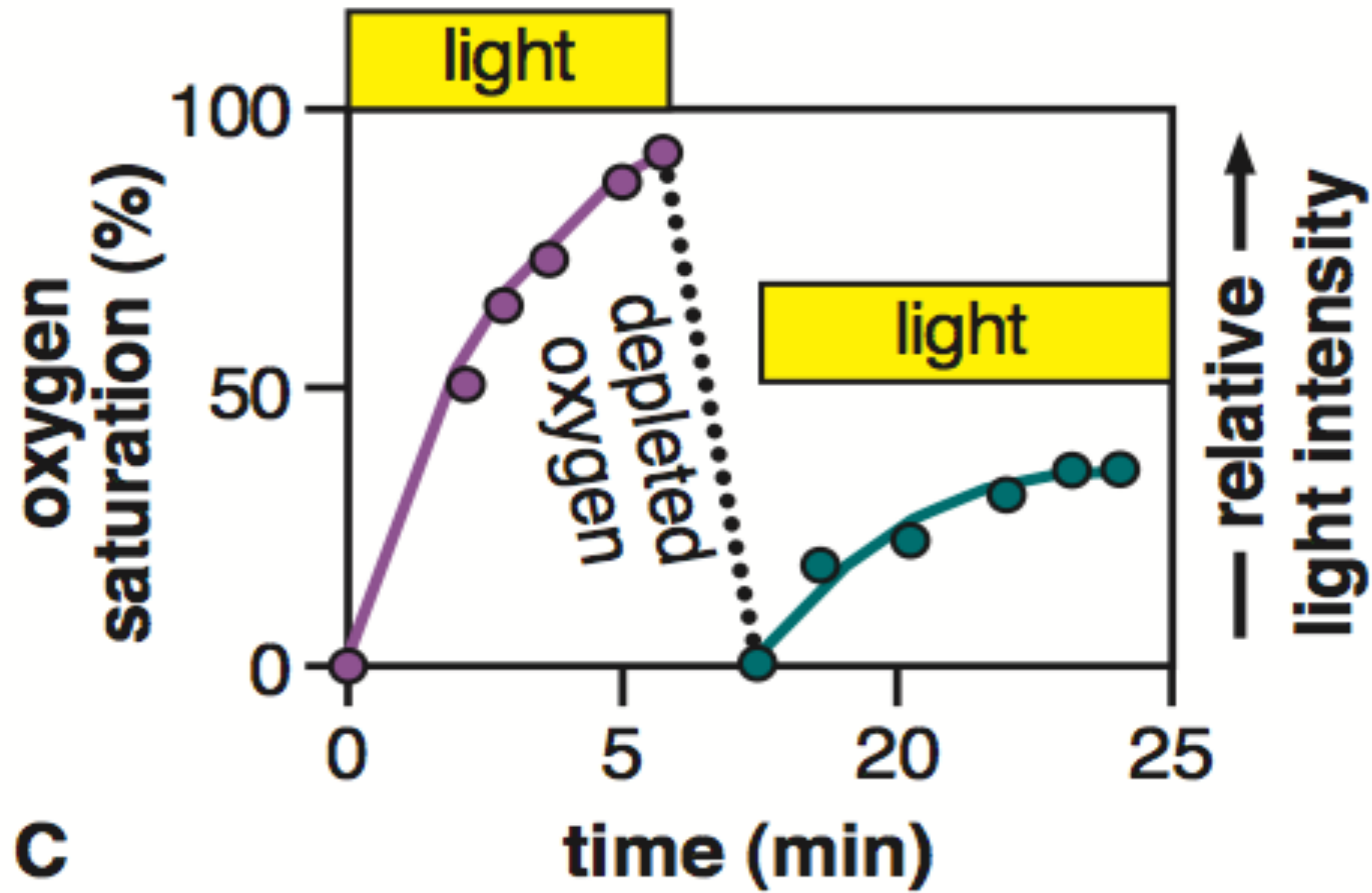


Fig. 11.2

# Influencing Oxygen Production Rate

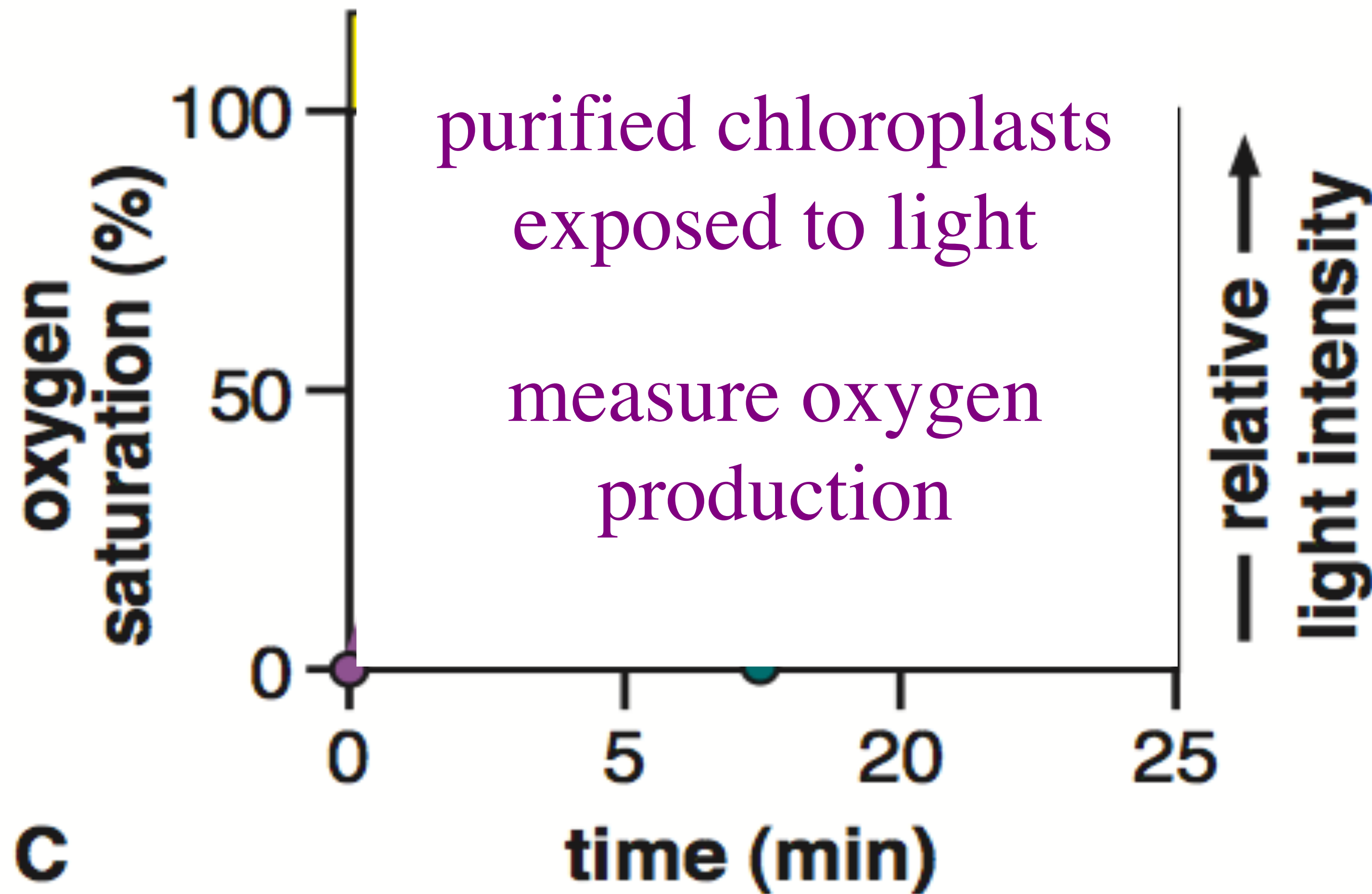


Fig. 11.2

# Influencing Oxygen Production Rate

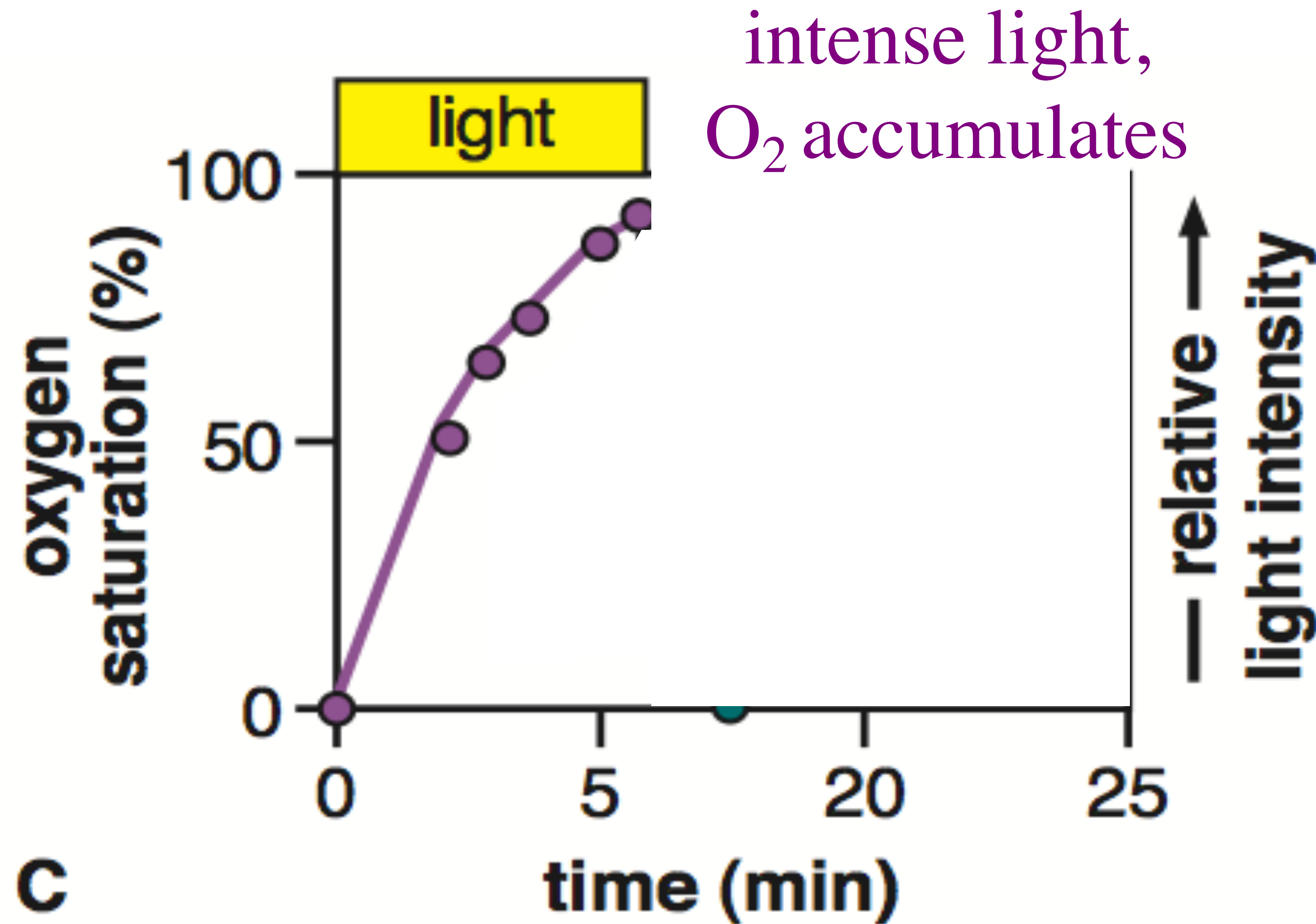


Fig. 11.2

# Influencing Oxygen Production Rate

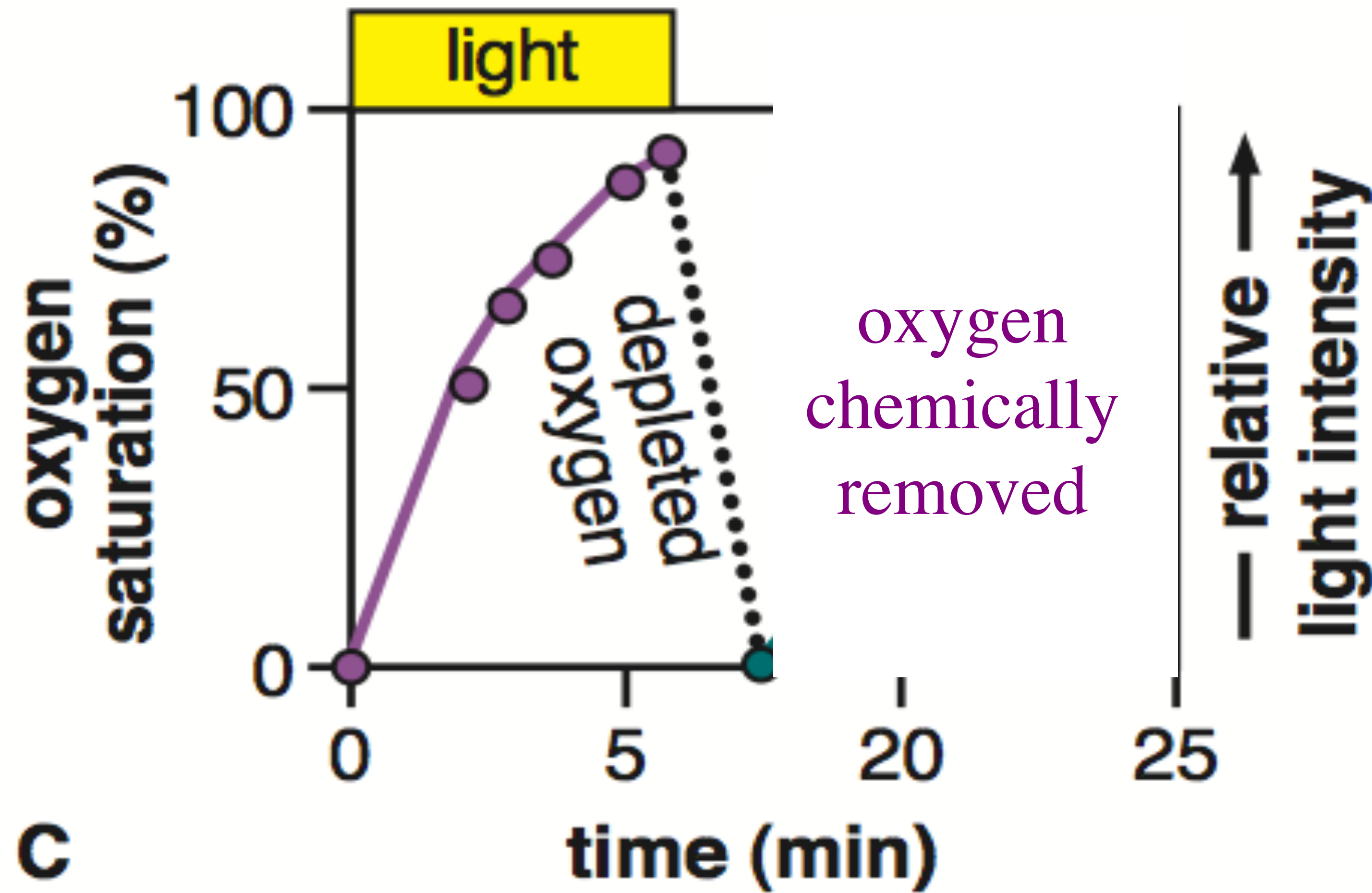


Fig. 11.2

# Influencing Oxygen Production Rate

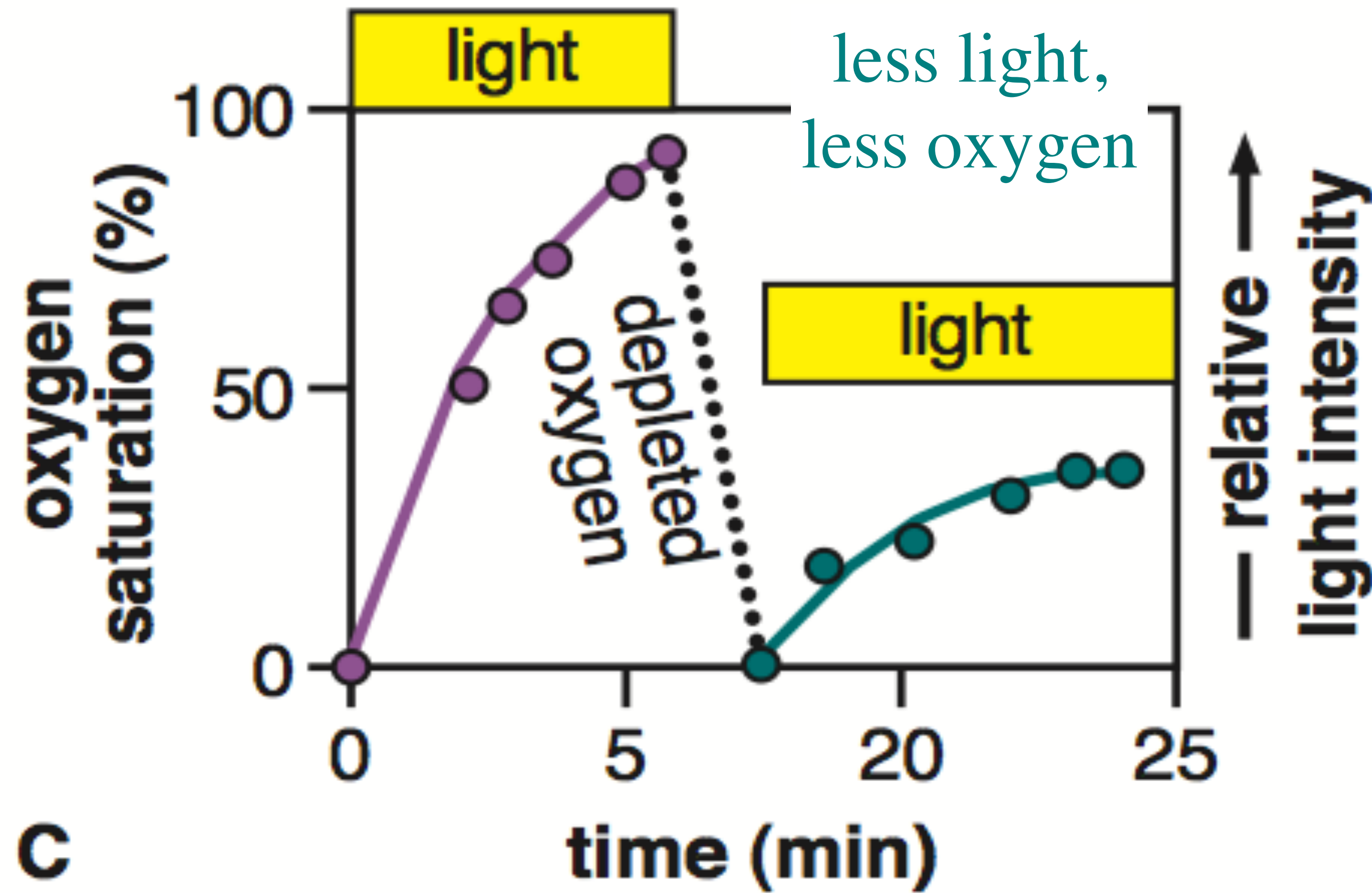
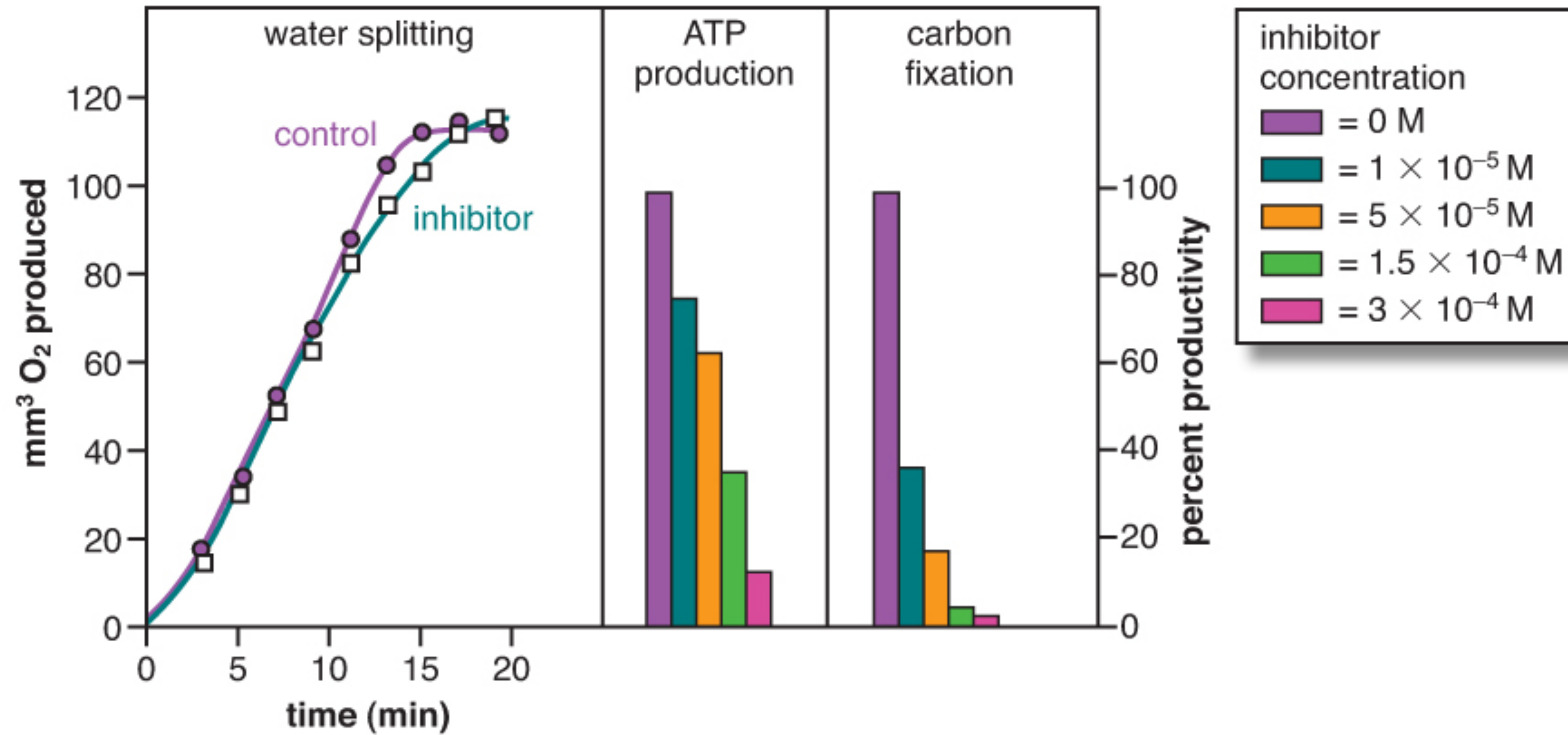


Fig. 11.2

# Trifecta



**Figure 11.4** Photosynthesis is a summation of three parts. Using an enzyme inhibitor, botanists dissected photosynthesis by measuring the production of oxygen ( $\pm 10^{-5}$  M inhibitor), ATP, and carbon fixation (at the indicated inhibitor concentrations). Modified from Arnon et al., 1954; their figure 1. Reprinted by permission from Macmillan Publishers Ltd: Nature. Photosynthesis by Isolated Chloroplasts. Vol. 174(4426). copyright 1954.

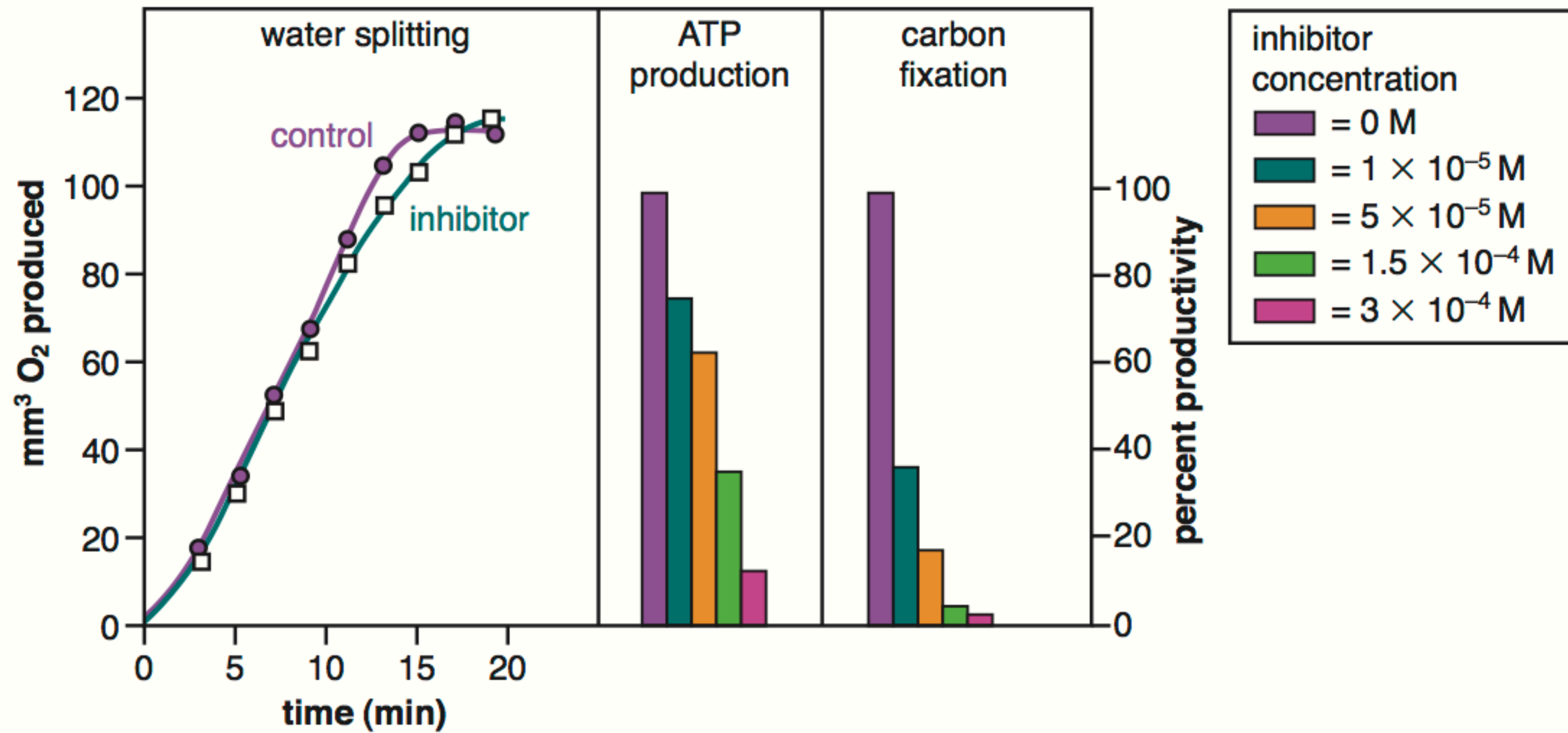


Fig. 11.4

modified from Arnon *et al.*, 1954

# Photosynthesis Composed of Steps

test 3 products separately

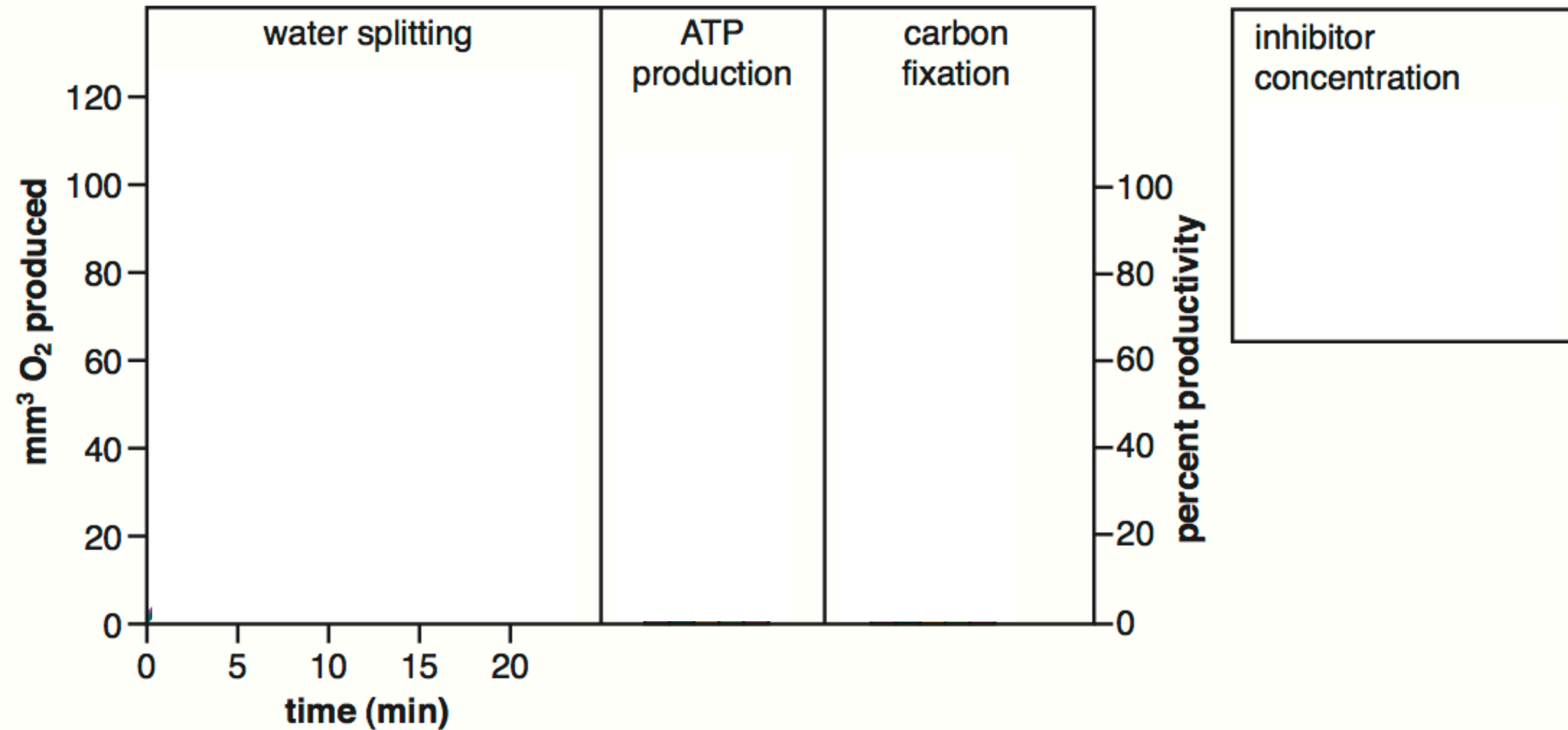


Fig. 11.4

modified from Arnon *et al.*, 1954

# Photosynthesis Composed of Steps

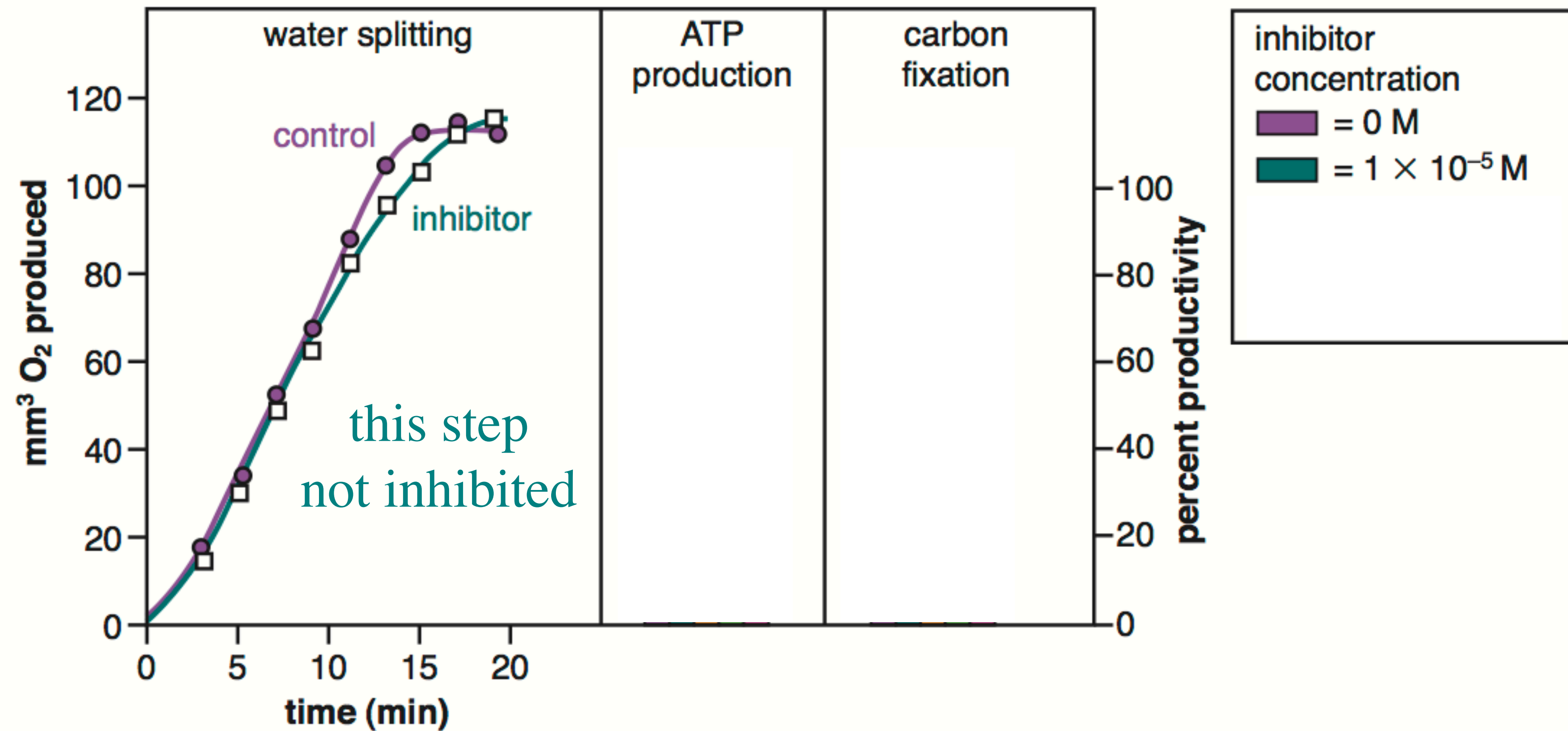


Fig. 11.4

modified from Arnon *et al.*, 1954

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# Photosynthesis Composed of Steps

ATP production  
partially inhibited

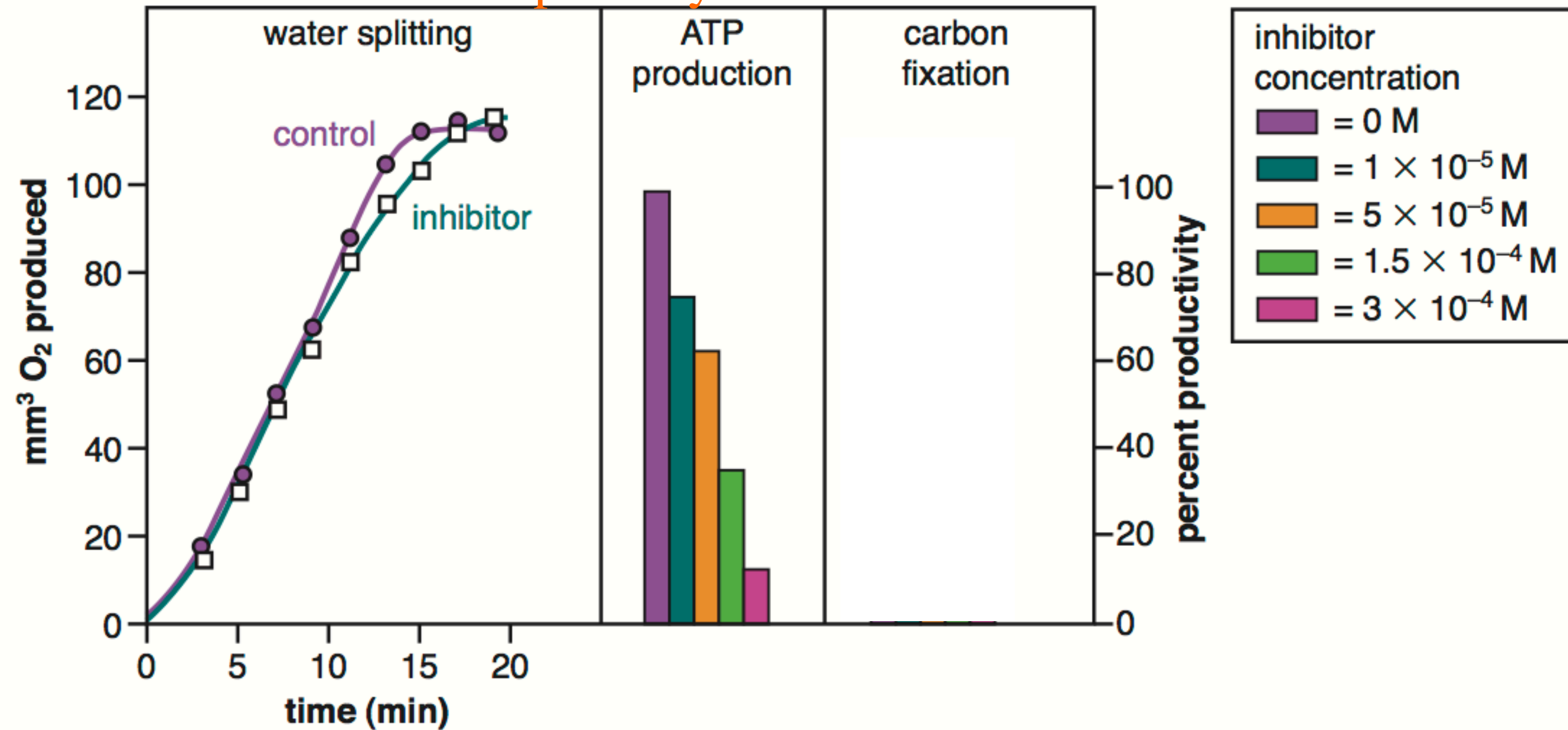


Fig. 11.4

modified from Arnon *et al.*, 1954

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# Photosynthesis Composed of Steps

CO<sub>2</sub> fixation  
inhibited faster

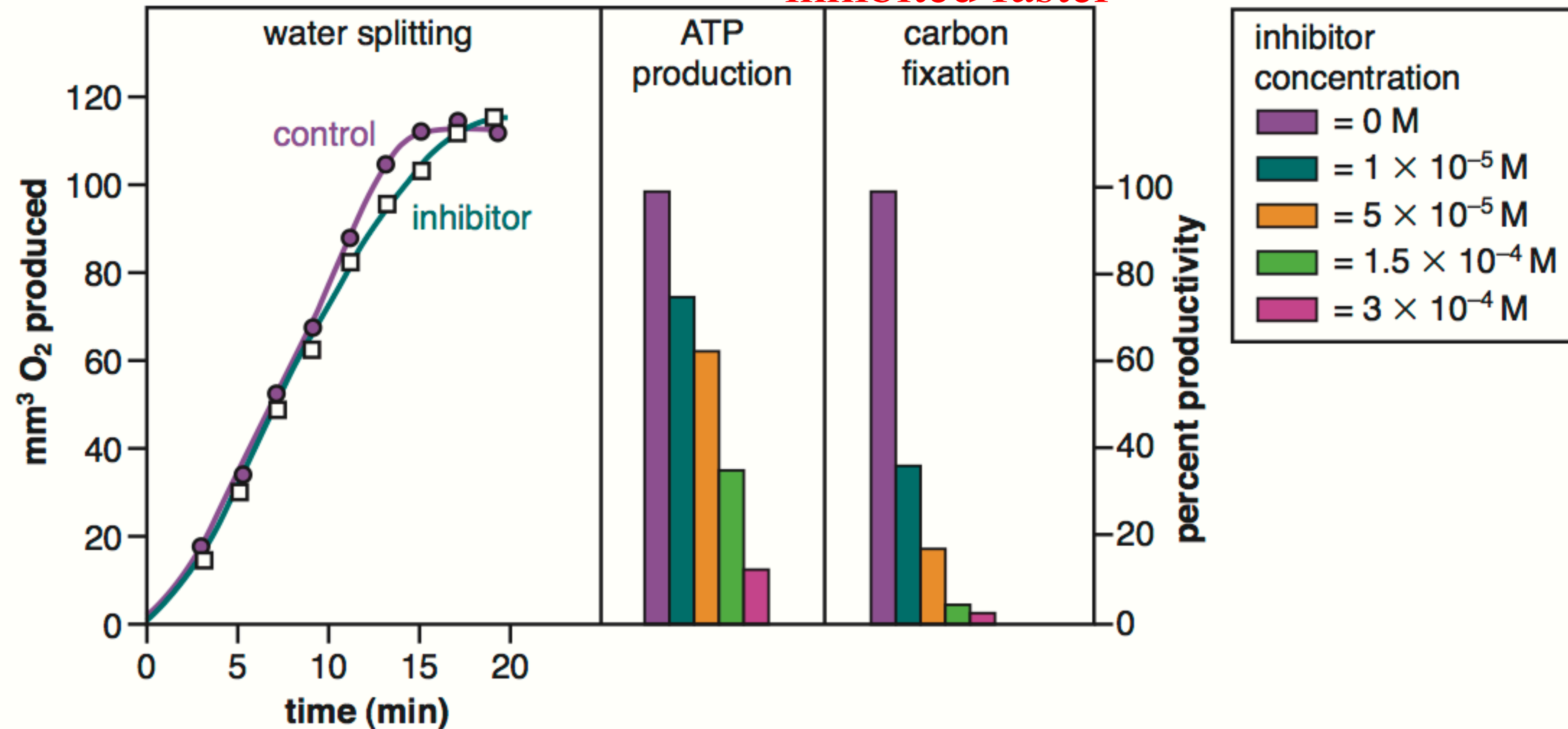
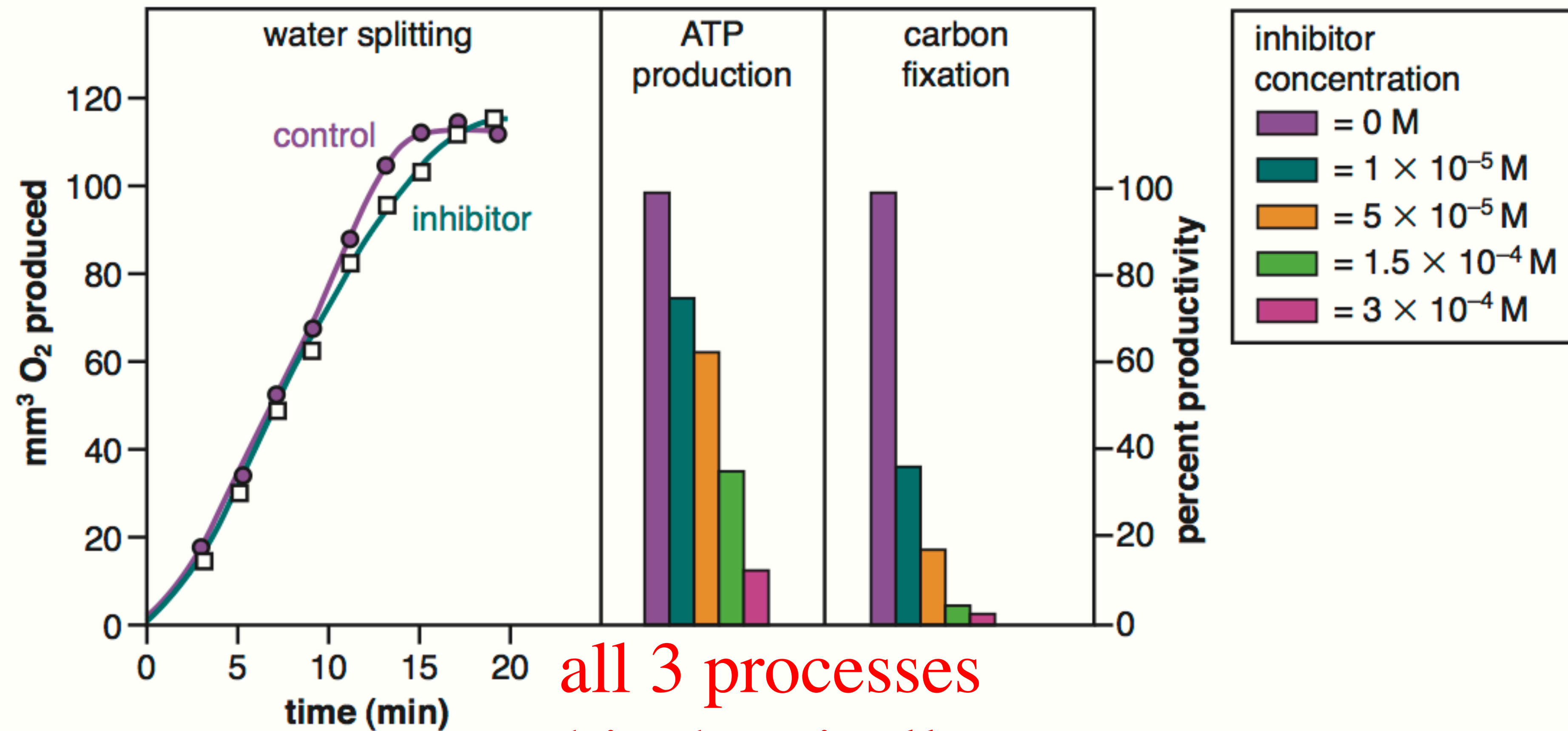


Fig. 11.4

modified from Arnon *et al.*, 1954

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# Photosynthesis Composed of Steps



all 3 processes  
biochemically  
separate

Fig. 11.4

modified from Arnon *et al.*, 1954

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# PHOTOSYNTHESIS BY ISOLATED CHLOROPLASTS

By PROF. DANIEL I. ARNON, DR. M. B. ALLEN,  
and DR. F. R. WHATLEY

Department of Plant Nutrition, University of California,  
Berkeley

THE many attempts to reconstruct photosynthetic events by systems less organized than whole green cells have met with success in only one area: the photolysis of water by chloroplast preparations resulting in the evolution of oxygen<sup>1</sup> (the Hill reaction). Evidence has now been obtained that whole chloroplasts, unaided by other cellular particles, have the ability to carry out two other photochemical reactions outside the living cell: (a) carbon dioxide fixation, and (b) photosynthetic phosphorylation, a term which we use for the conversion of light energy into the high-energy phosphate bonds of adenosine triphosphate (ATP), without the participation of respiration.

Although there are valid reasons for believing that the functioning photochemical mechanism in isolated chloroplasts, as measured by the evolution of oxygen, is the same as in the intact cell<sup>2</sup>, there is uncertainty as to the manner in which light energy captured by chloroplasts is linked to carbon dioxide reduction. Until now, isolated chloroplasts appeared to differ

oxylation of pyruvate to malate, was in the cytoplasmic fluid<sup>4,9,10</sup>. Other carboxylases<sup>10</sup> as well as an important enzyme of the glycolytic cycle, glyceraldehyde phosphate dehydrogenase<sup>11</sup>, were also found in the cytoplasmic fluid of green leaves rather than in the chloroplasts.

Our present findings, however, speak against this concept. Since washed chloroplasts were found to have the capacity, without the addition of other enzymes, to use light energy for adenosine triphosphate synthesis and carbon dioxide fixation, it follows that these particles contain the enzyme systems and at least most of the co-factors concerned in these reactions. The only added compound which gave consistently large increases in both photosynthetic phosphorylation and carbon dioxide fixation was ascorbic acid. The addition of di- or triphosphopyridine nucleotide, uridine triphosphate, cocarboxylase, thiocetic acid or a mixture of thiocetic acid and cocarboxylase was without effect on carbon dioxide fixation or photosynthetic phosphorylation. In the light of our present evidence, isolated chloroplasts emerge as remarkably complete cytoplasmic structures, equipped to carry out not only oxygen evolution but also carbon dioxide fixation and the conversion of light into chemical energy.

In setting out to investigate light reactions of isolated chloroplasts with reference to photosynthesis, our basic premise has been that only those chloroplast preparations which possessed the capacity for photolysis of water, as measured by oxygen evolution, were desirable objects of study. Isolated chloroplasts lacking this capacity are, so far as we know, incapable

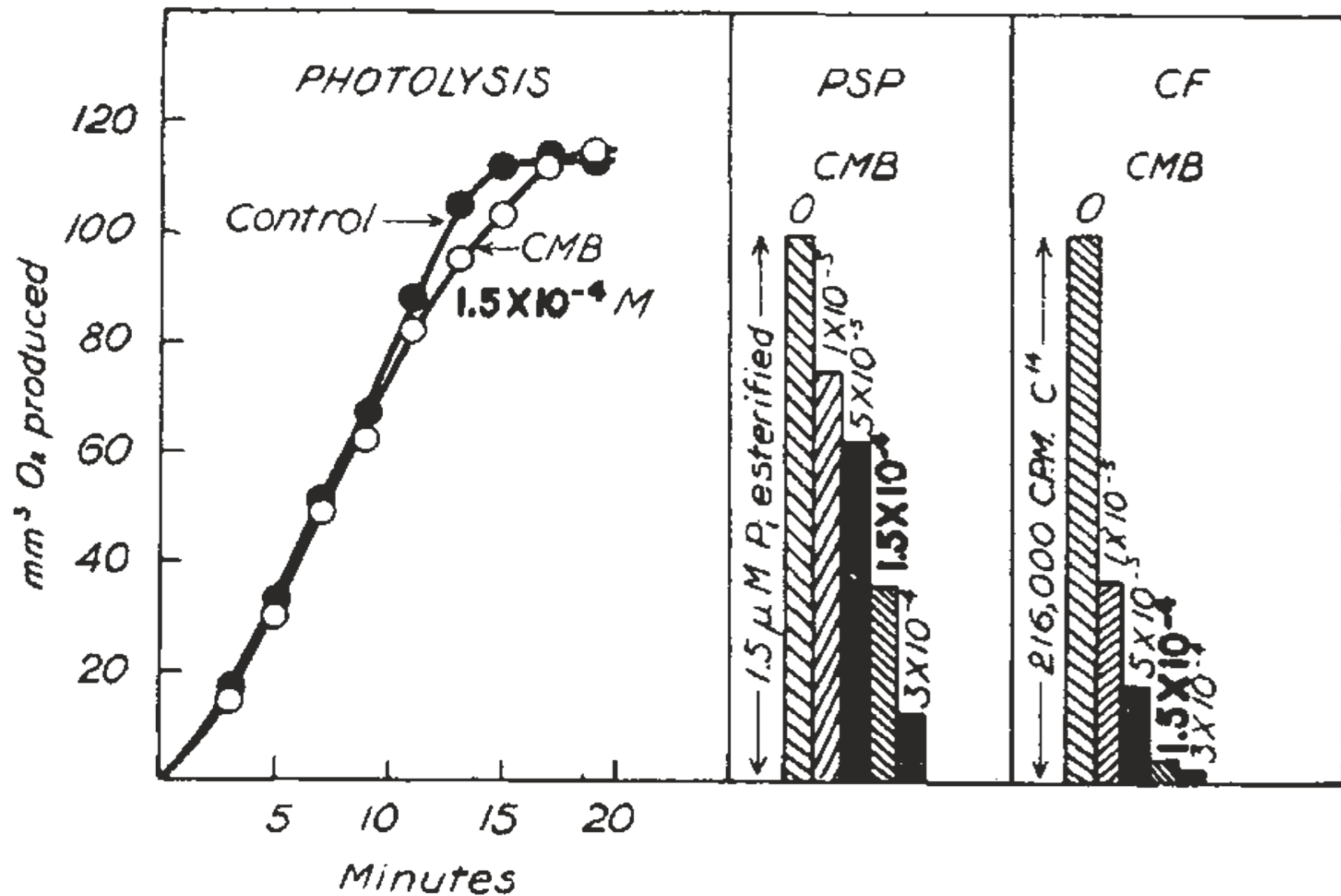
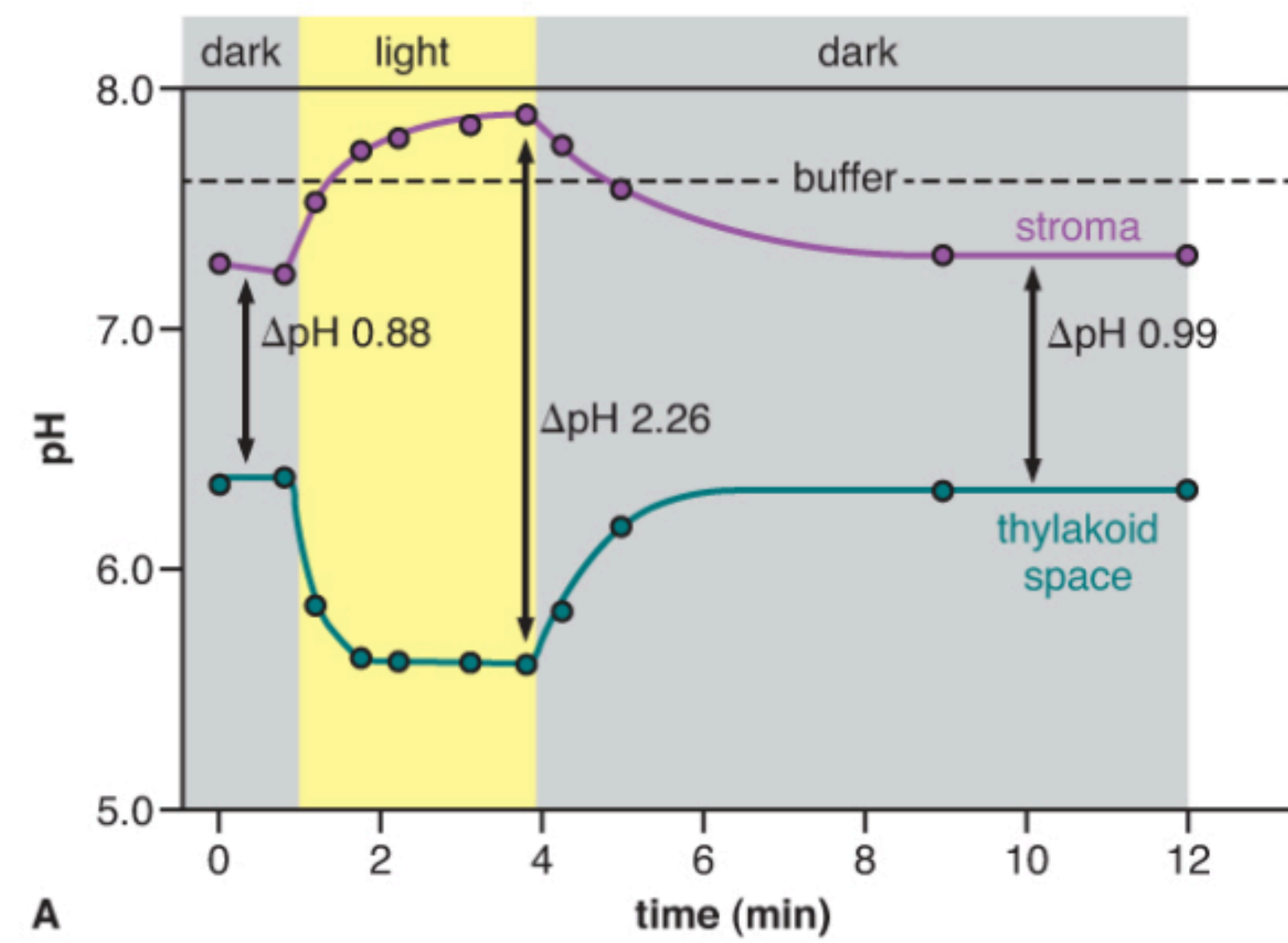
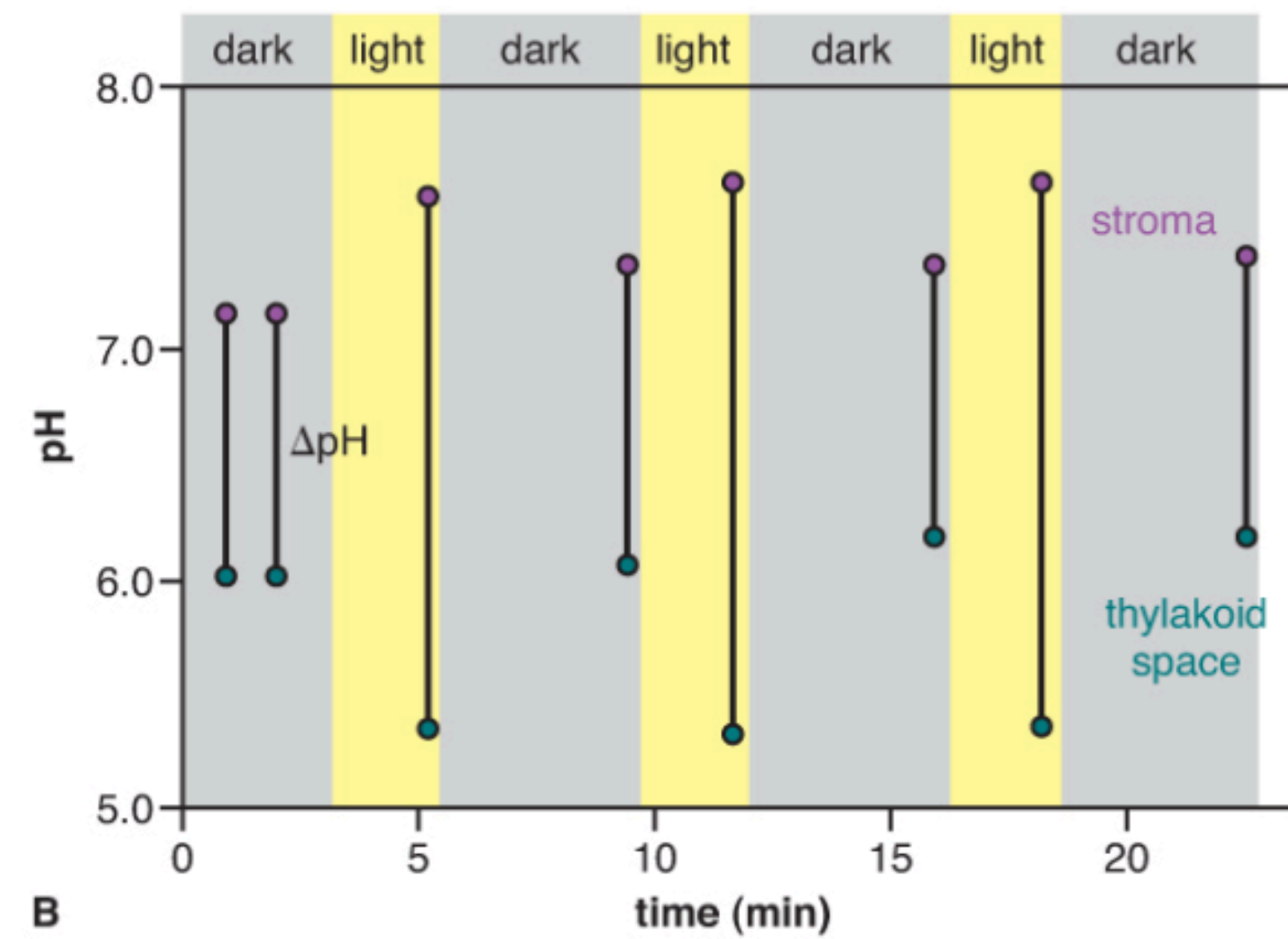


Fig. 1. Effect of *p*-chloromercuribenzoate (CMB) on photolysis, photosynthetic phosphorylation (PSP) and carbon dioxide fixation (CF) by isolated chloroplasts. Experimental conditions as in Table 1, except that gas phase in photolysis and carbon dioxide

# Trifecta



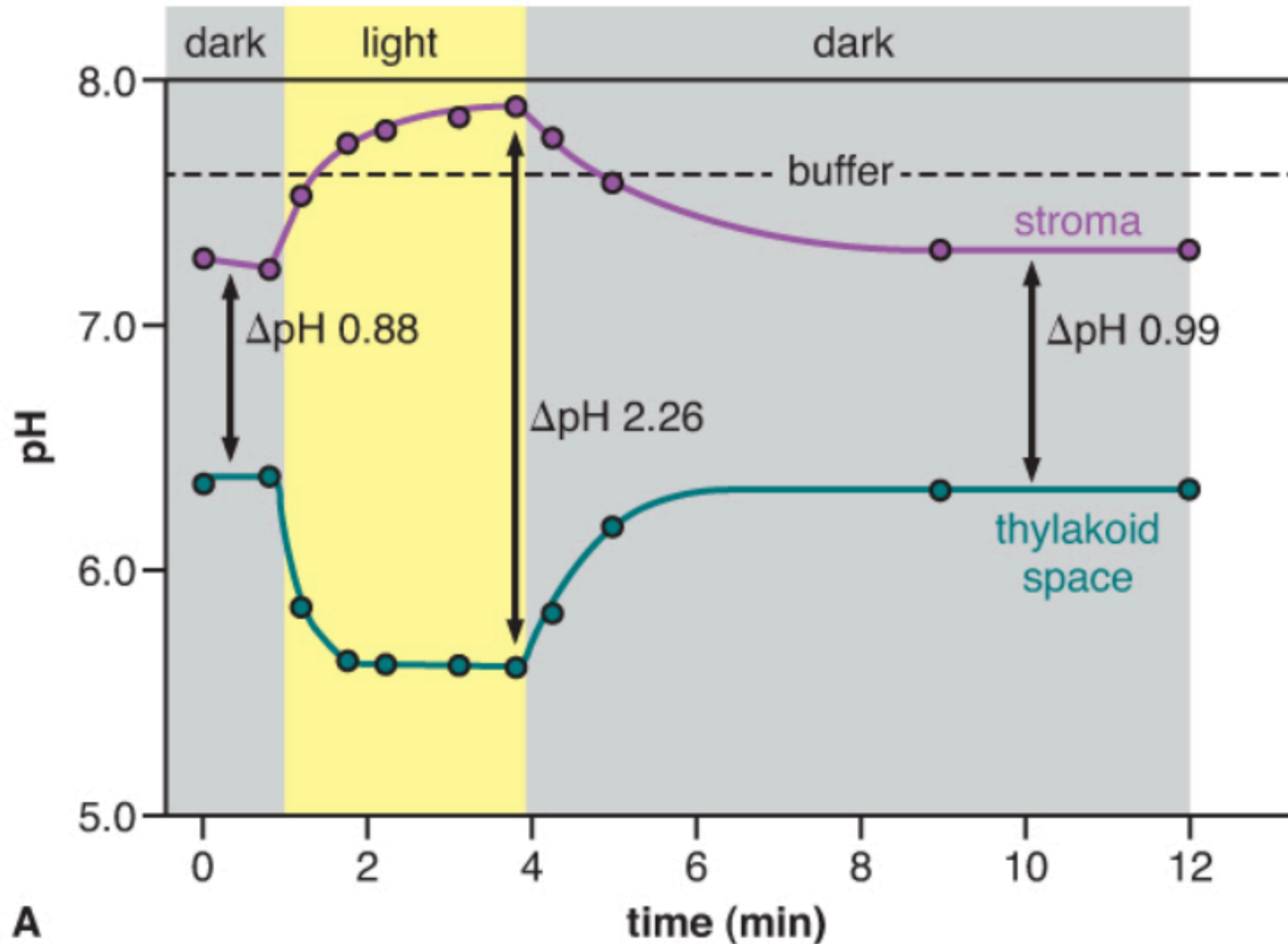
A



B

**Figure 11.5** Comparison of pH in stroma and thylakoid space. **A**, Botanists measured the pH of a single chloroplast exposed to dark or light, as indicated. **B**, Repeated exposures of a chloroplast to light and dark cycles within minutes of each other. Panel A modified from Heldt *et al.*, 1973; their figure 4. Panel B modified from Heldt *et al.*, 1973; their figure 5. Heldt, Hans W., Karl Werdan, *et al.*

# Trifecta



# pH Differential in Chloroplast

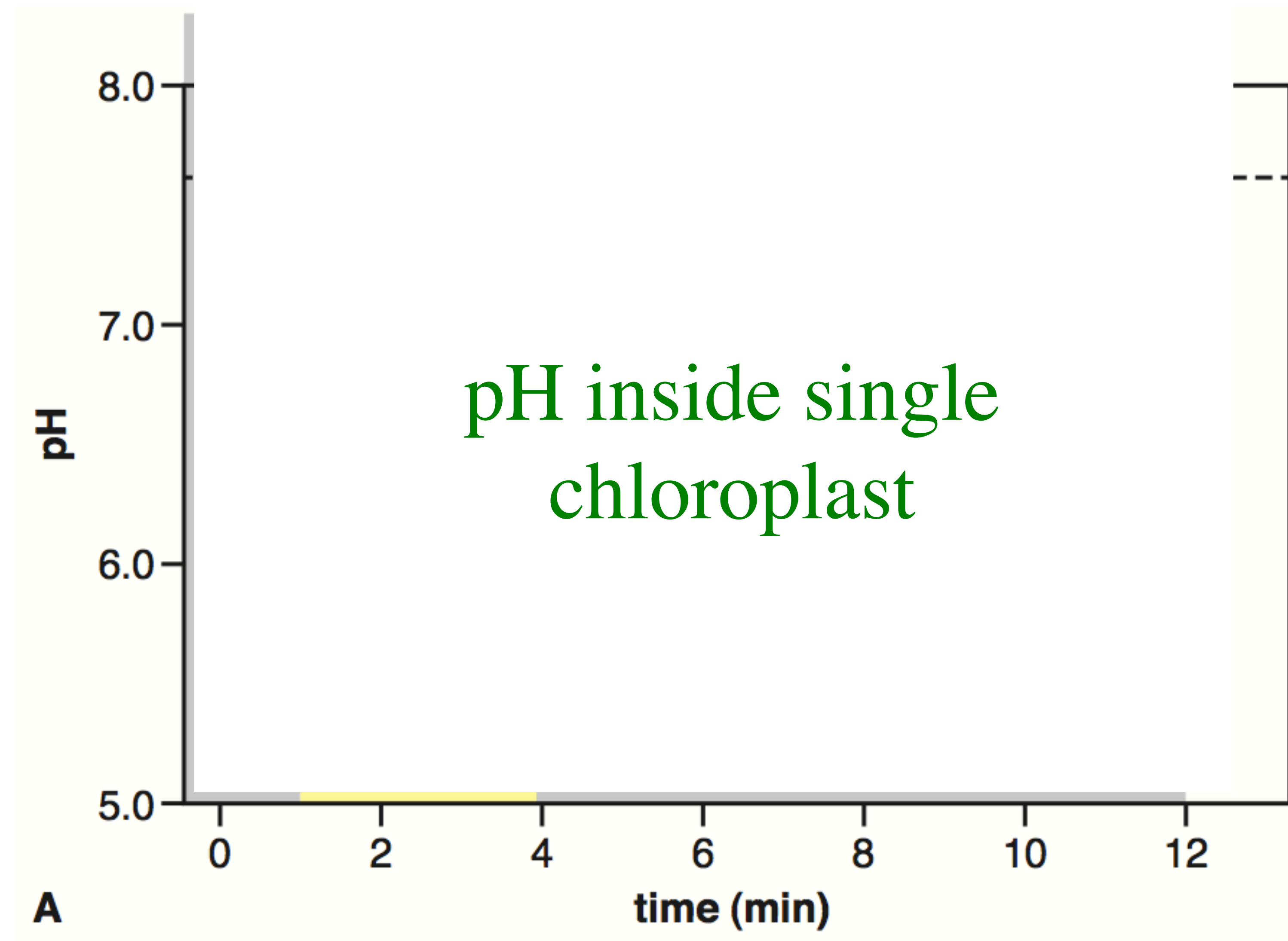


Fig. 11.5

# pH Differential in Chloroplast

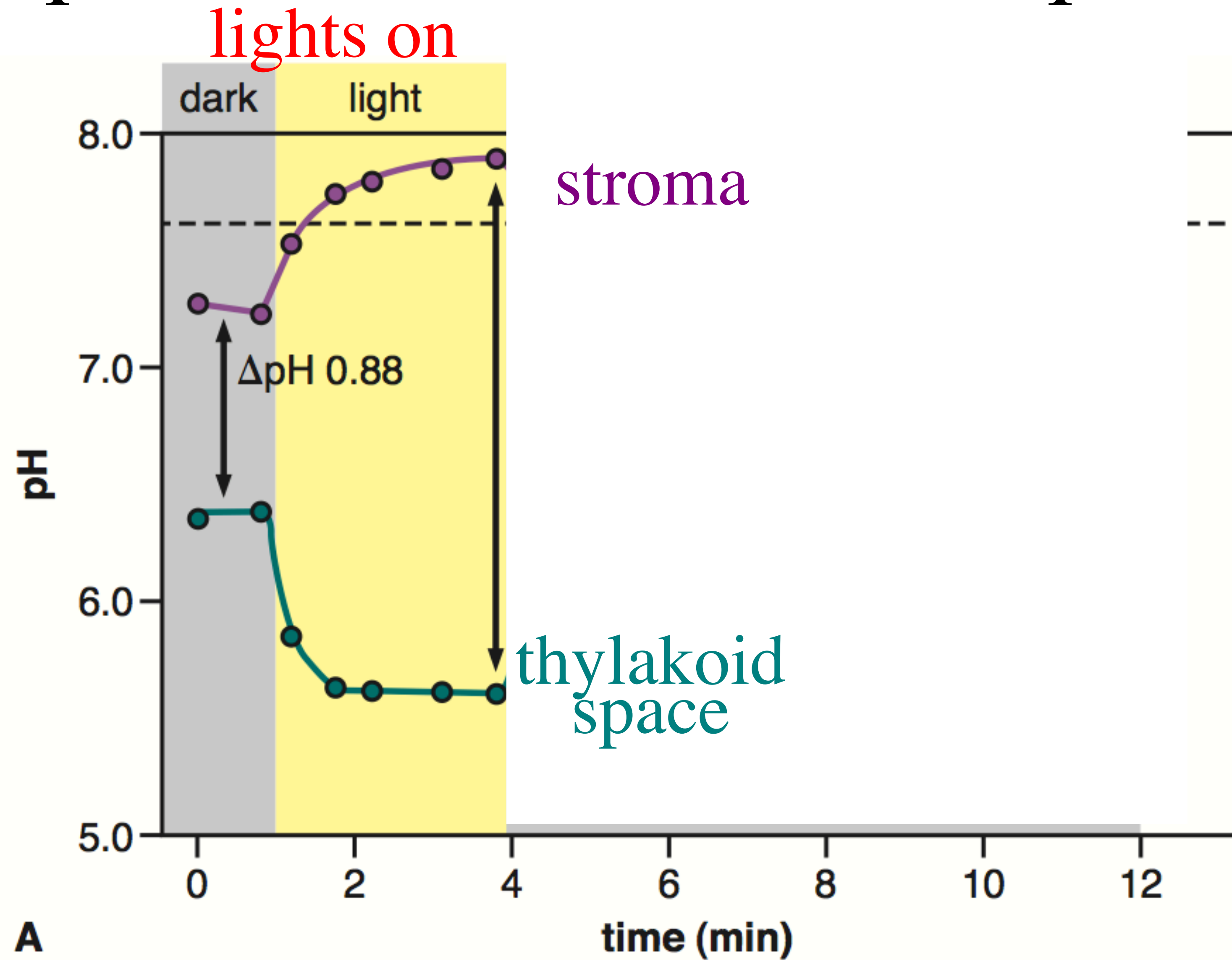
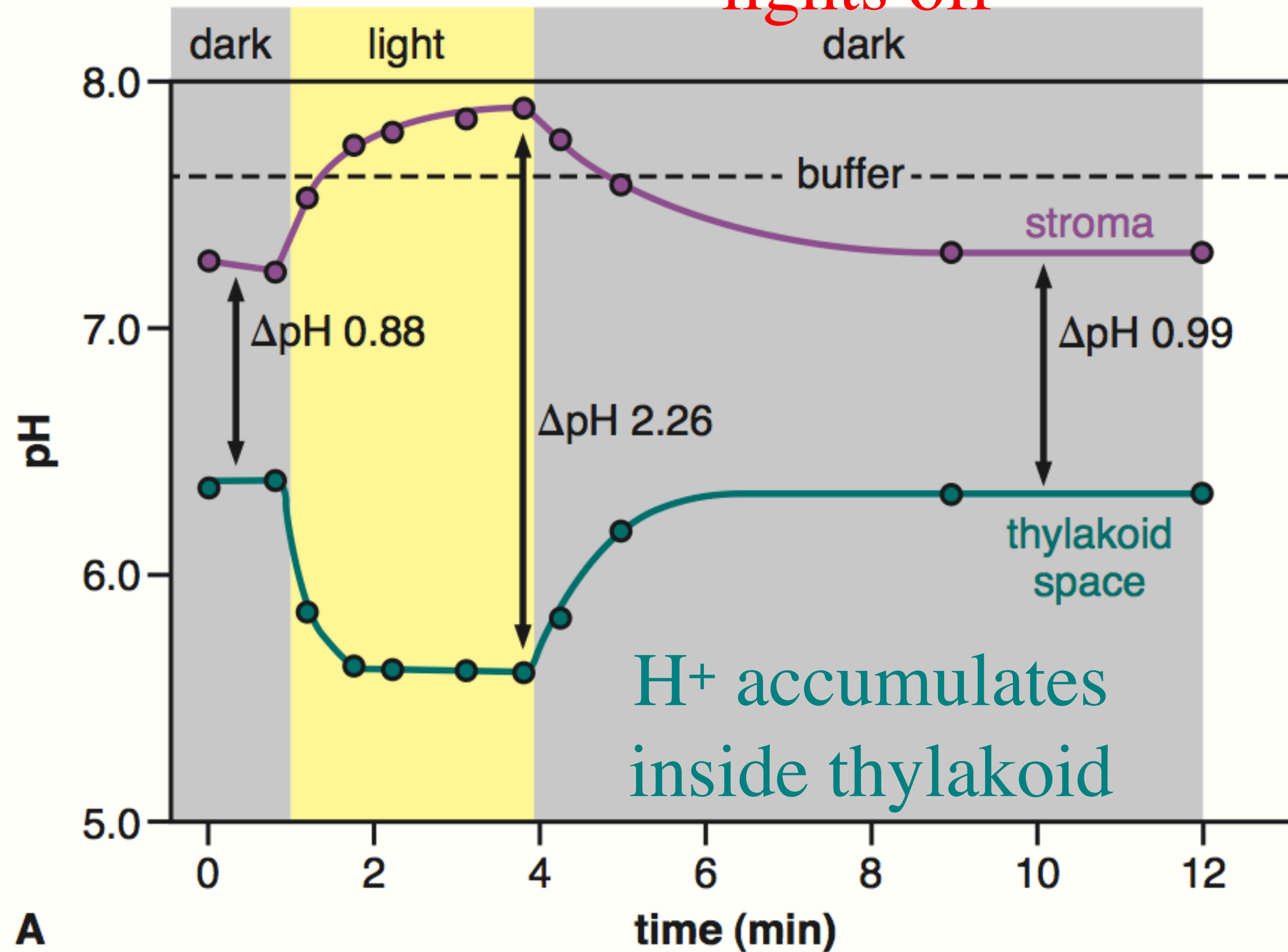


Fig. 11.5

# pH Differential in Chloroplast

lights off



A

Fig. 11.5

# pH Differential in Chloroplast

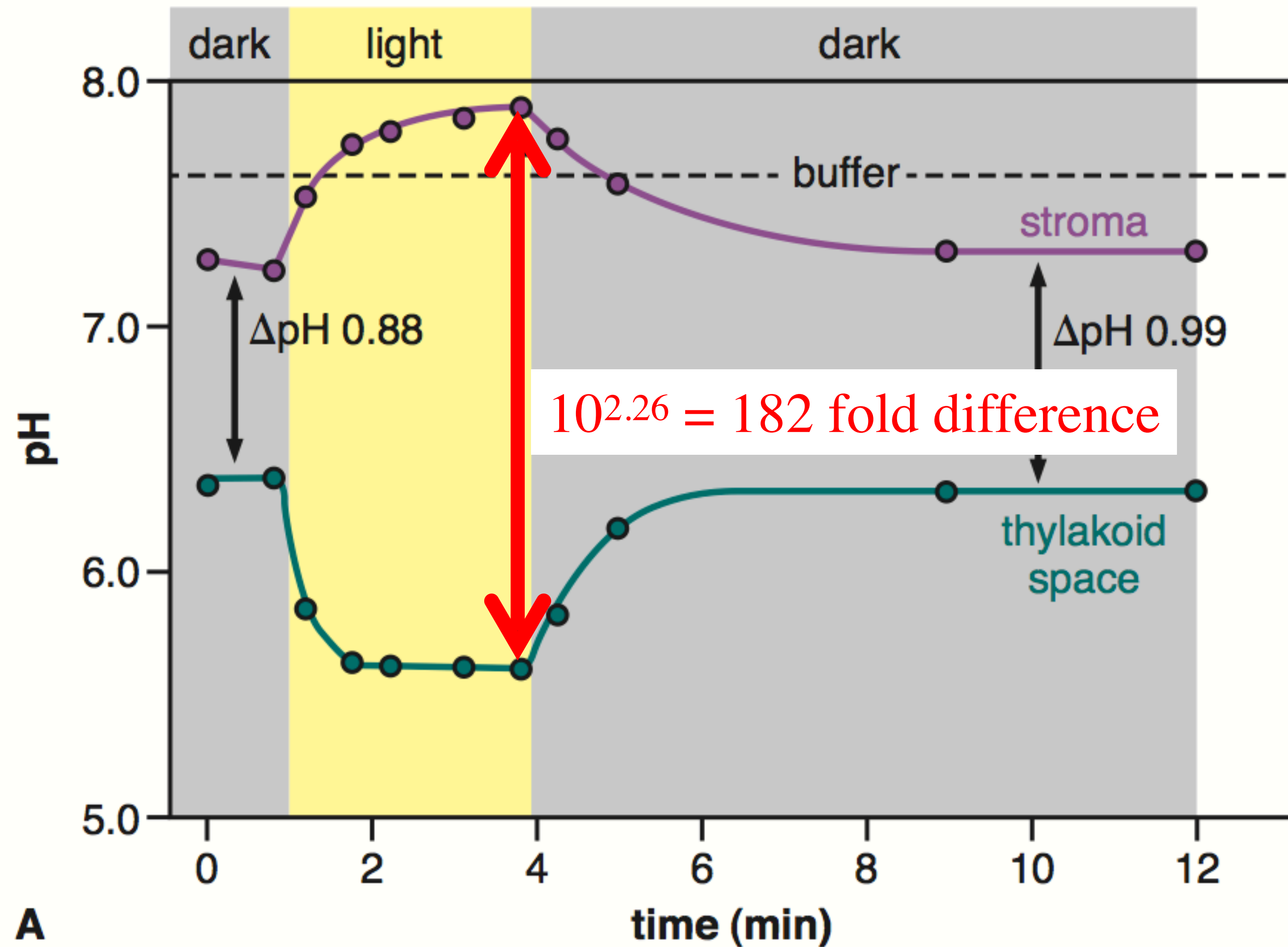
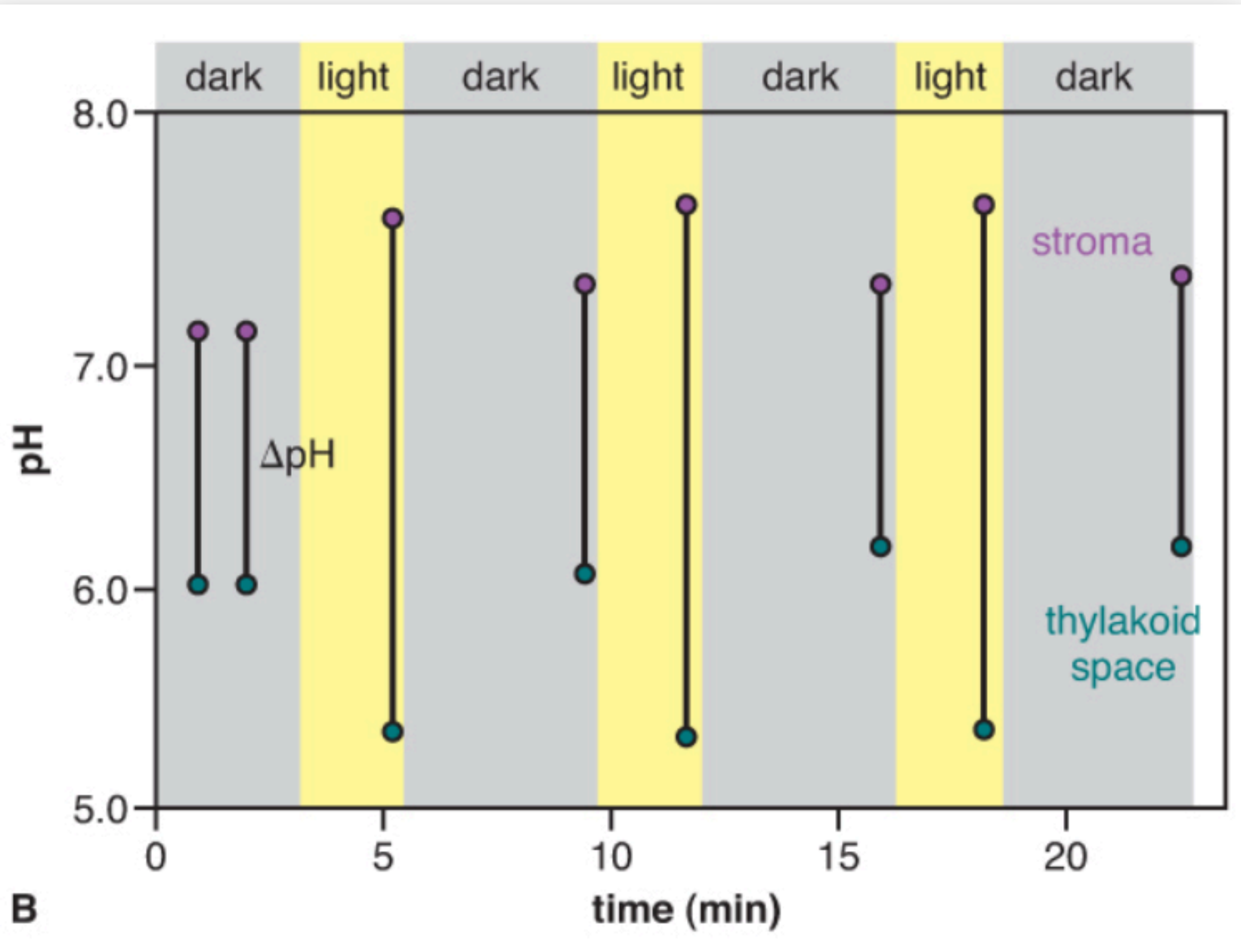


Fig. 11.5

# Trifecta



**B**

# Can Chloroplast Recharge & Repeat?

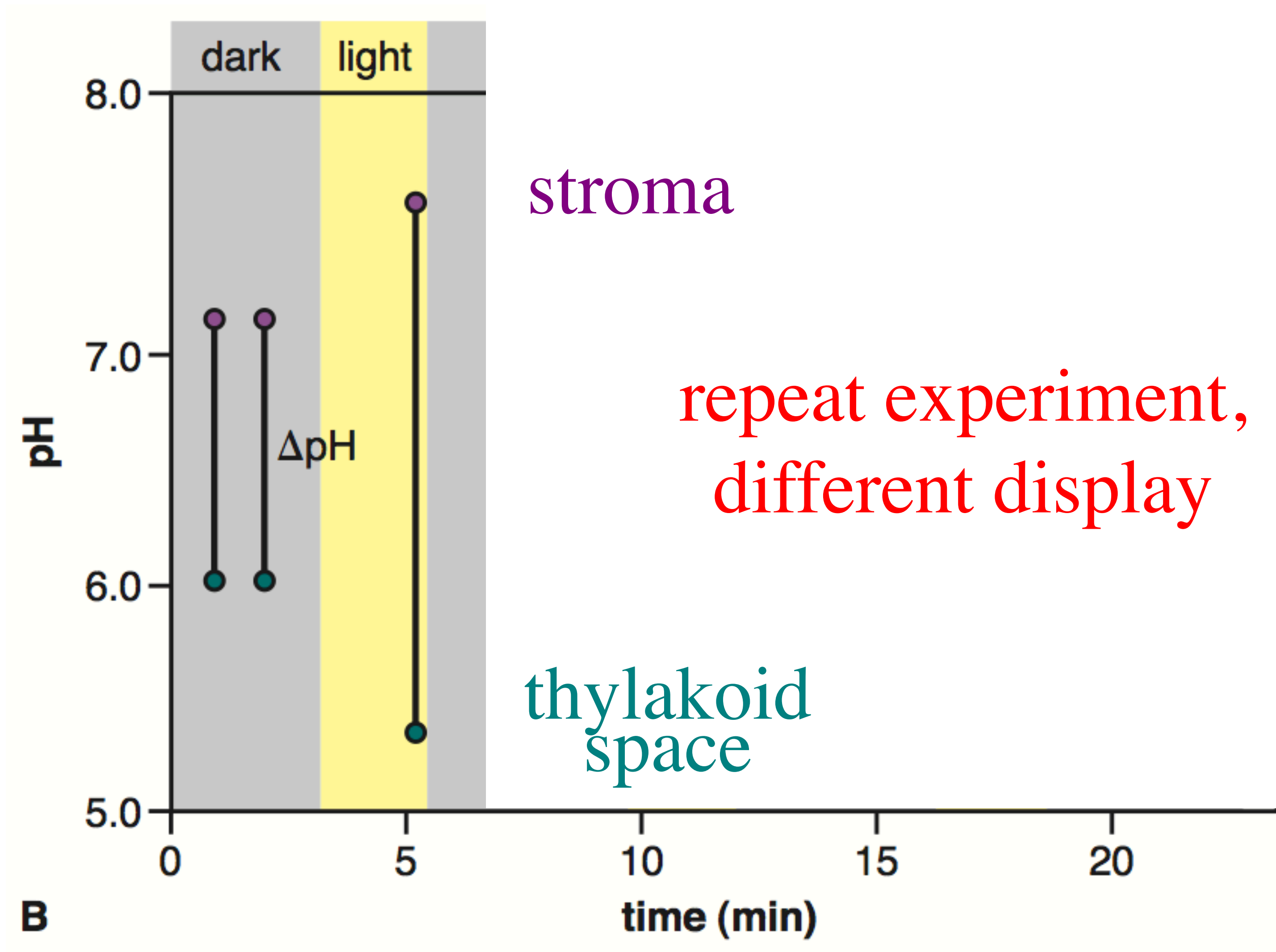


Fig. 11.5

# Can Chloroplast Recharge & Repeat?

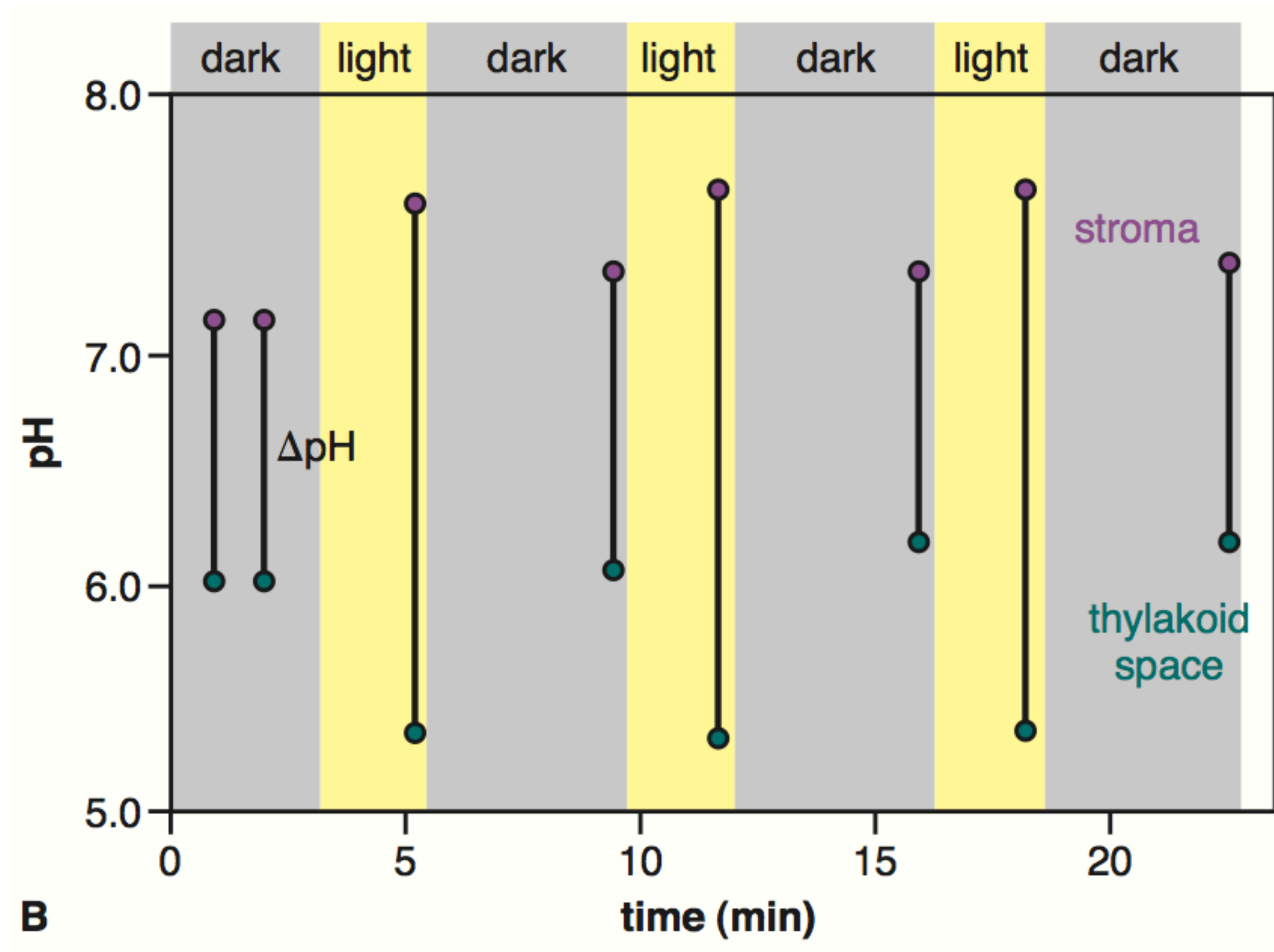


Fig. 11.5

BBA 46590

## ALKALIZATION OF THE CHLOROPLAST STROMA CAUSED BY LIGHT-DEPENDENT PROTON FLUX INTO THE THYLAKOID SPACE\*

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(Received March 19th, 1973)

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### SUMMARY

1. From the uptake of dimethyloxazolidinedione and of methylamine into the sucrose-impermeable space of intact spinach chloroplasts as measured by silicon-layer filtering centrifugation and from estimation of the size of the thylakoid space by planimetry of electron micrographs the pH in the stroma and in the thylakoid space is evaluated. The reliability of the method is checked.

2. Illumination causes an alkalization in the stroma and an acidification in the thylakoid space, with a  $\Delta\text{pH}$  of about 2.5 between the two spaces, reflecting light-

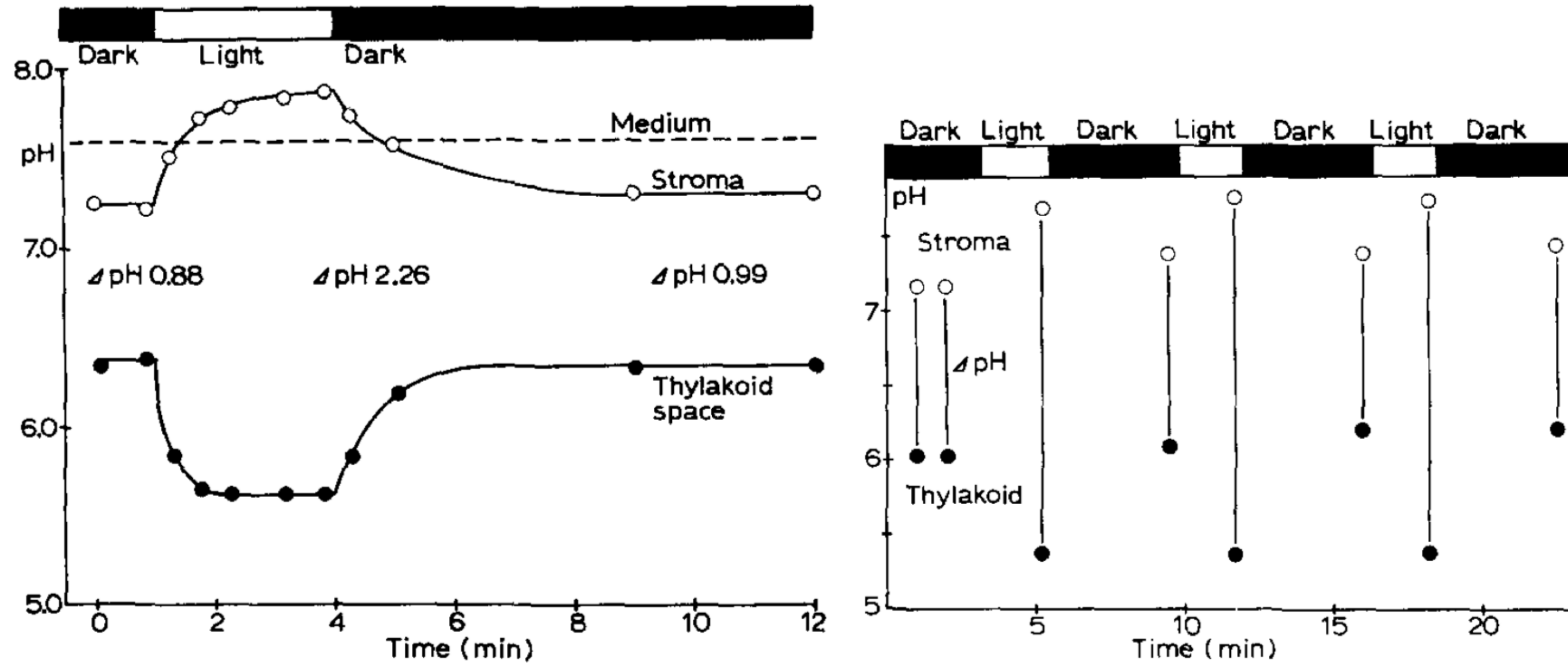


Fig. 4. Changes of the pH in the stroma and in the thylakoid space of spinach chloroplasts (45.0 μg chlorophyll/ml) caused by dark–light and light–dark transients. Temp., 8 °C. Sucrose-impermeable <sup>3</sup>HHO space, 26.3 μl/mg chlorophyll. Each sample has been incubated in the centrifugation tube and the incubation was terminated by centrifugation. For details see Methods.

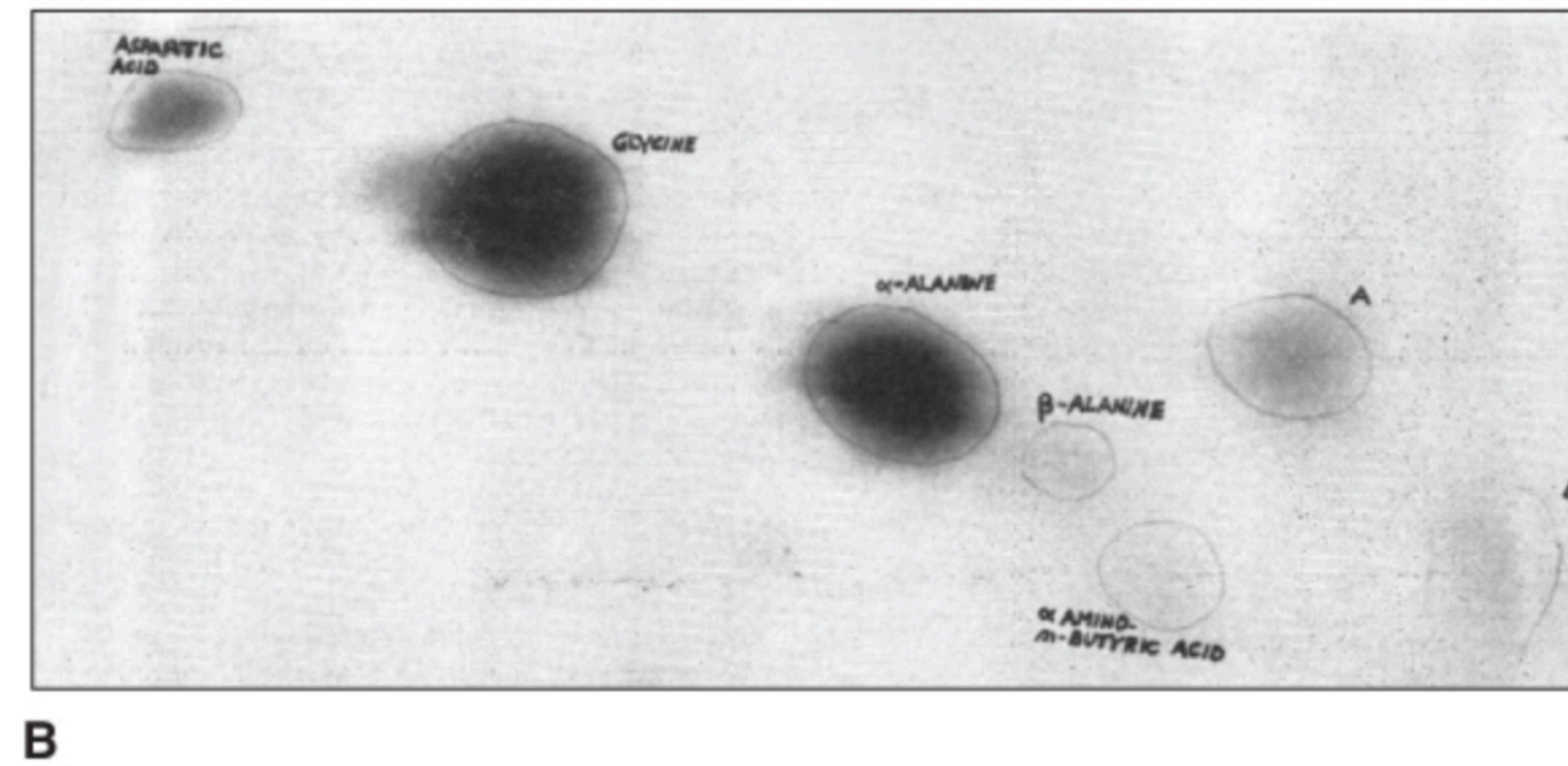
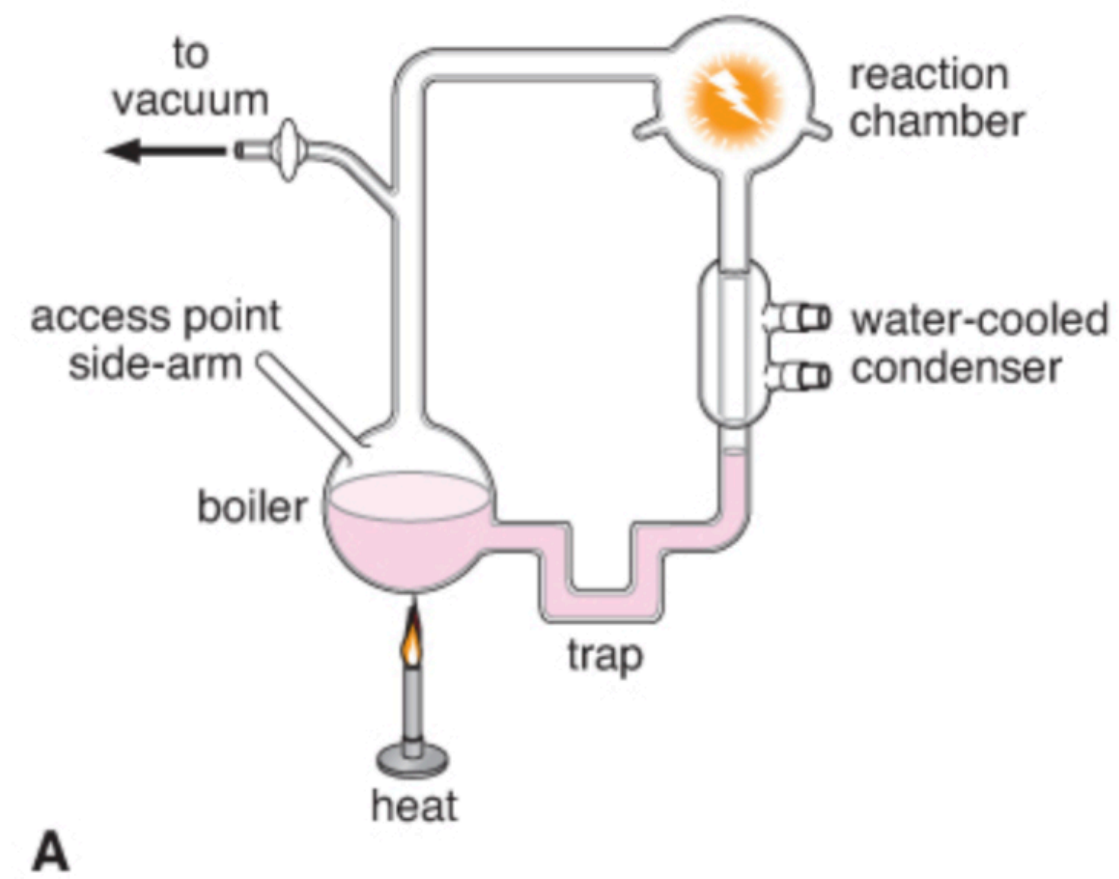
Fig. 5. Change of the pH in the stroma and in the thylakoid space of spinach chloroplasts (57.0 μg chlorophyll/ml) caused by repetitive dark–light and light–dark transients. For details see Methods and legend Fig. 4. Sucrose-impermeable <sup>3</sup>HHO space: light, 21.3 μl/mg chlorophyll; dark, 21.9 μl/mg chlorophyll. ○, pH in the stroma space; ●, pH in the thylakoid space.



## Possible exam question format

Predict how many photons of red light are minimally required to create one glucose molecule in photosynthesis (or two glyceraldehyde-3-phosphate molecules) and explain your reasoning. (i) **Illustrate** the non-cyclic electron transport version of light reactions and the Calvin cycle. (ii) **Explain** in full sentences your prediction and your rationale for it. Explain in detail the sequence of events that occur in the process of photosynthesis and your step-by-step rationale/logic in your calculations [Assume: the Calvin cycle must occur in full cycles, you must only use linear electron transport, 3 H<sup>+</sup> travel through ATP Synthase for it to generate one ATP].

## Possible exam question format



6) What was the outcome of this experiment on the origin of life? Structure your answer into: Purpose, Methods, Findings.

## Possible exam question format



5) Explain the Purpose, Methods and Findings of Stanley Miller's famous experiment. (50 word limit)

## Possible exam question format

**What is a multiple True/False/Why question?**

(Multiple-choice with [partial credit!](#))

Example:

**1. What are the official sport team colors of Michigan State University?**

a. Maize

b. Blue

c. Green

d. Yellow

e. White

**f. Why?:** Historically, why were those colors chosen?

For full credit you must respond with:

The answer "c" is True

The answer "e" is True

To answer to "f" you accurately explain why those are the colors

## How should I study ?

### Dr Lynn Margulis

- Who is she, what's the purpose of her research, and relevance?
- Draw a figure from her work and Trifecta it.
- Explain two learning goals associated with her work. What are you supposed to learn as a result?

*Yet remember: Can bring 3x5 inch card with handwritten notes on both sides.*