

Lecture 6: Pigments and Photosynthesis

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1 Content

- Discuss the importance of light in photosynthesis and the different types of pigments involved.
- Explain the concept of light compensation point and light saturation point.

2 Aims

The lecture will focus on the importance of light for photosynthesis, exploring the different types of pigments involved and physiological concepts such as the light compensation point and light saturation point.

2.1 Lecture transcript

2.1.a Part I

Welcome back again to BDC 223. Today we're going to be talking about the development of the theory of chromatic adaptation. This is an interesting story that tells us a bit about how science works and shows that over about 150 to 170 years, our understanding of how plants become adapted to variable light environments (variability in both quality and quantity of light) has changed through the accumulation of different kinds of evidence and through technological advances that have made various measurements possible. It also changed as a result of the accumulation of empirical evidence over many years.

Let's look at some of the different contributors to our theories of chromatic adaptation. It starts at the beginning with Theodore Engelmann, who published his theories of chromatic adaptation around 1881 to 1883. He based much of his thinking on where seaweeds live in the ocean and previous work from 1843 by Anders Ørsted. Ørsted observed that seaweeds come in different colours: green algae, brown algae, and red algae (Chlorophyta, Phaeophyta, and Rhodophyta, respectively). Ørsted suggested that perhaps these colours correspond to adaptation to different places in the ocean.

All of this work about Anders Ørsted and Theodore Engelmann is discussed in detail in the paper by Mary Beth Saffo, “New Light on Seaweeds.” Please read that paper carefully. Everything you need to know about the variability of light in the ocean (the different kinds of pigments, light quantity, and light quality) is discussed in great detail. She uses it to develop the storyline of how chromatic adaptation developed over roughly a century.

Up until about 1980 or 1990, especially zoologists, tended to believe that chromatic adaptation was a real phenomenon. Spoiler alert: despite making sense on paper and passing various thought processes, it didn’t pan out precisely as originally thought. We’ll work through all these lines of evidence in the next few lectures.

Engelmann took note of Ørsted’s observation that algae come in red, brown, and green. He noticed that light penetrates the ocean differently: shallow coastal waters have red and blue light, intermediate levels have green light, and at greater depths, only dim blue light remains. Engelmann hypothesised that the distribution of seaweed colours vertically in the ocean could be explained by light quality—i.e., the colour of light affected which seaweeds were found at different depths.

He hypothesised that green seaweeds, being able to absorb red and blue light well, would be most abundant in shallow water. For brown seaweeds, able to absorb green light effectively, he expected their abundance in intermediate ocean depths. He believed red algae, which absorb blue light, would be found in deeper regions where blue light is more available. This seemed like a reasonable hypothesis and is something people might propose even today.

To test this, Engelmann designed a clever experiment using a prism made by Carl Zeiss, the German father of optics whose company still makes lenses, cameras, and microscopes today. Engelmann’s experiment used both macro- and unicellular algae, such as *Cladophora capensis* (a green alga), diatoms (golden brown algae due to their xanthophyll pigments), and *Polysiphonia* sp. (a filamentous red algae with phycobilin pigments). Each group of algae has its characteristic pigments: green algae with chlorophyll-*a* and -*b*, brown algae with xanthophylls, and red algae with phycocyanin and phycoerythrin.

Engelmann exposed these algae to split light from the Zeiss prism across a microscope slide, with red light at one end and blue at the other. He added aerotactic bacteria (bacteria attracted to oxygen) to the watery medium around the algae. The idea was: the colour of light driving the most photosynthesis would result in the most oxygen production, so the bacteria would accumulate in those regions.

He found that bacteria clustered in regions where the predominant pigment of the algae produced the most oxygen. For *Cladophora* with chlorophyll-*a*, bacteria accumulated where chlorophyll-*a* absorbs best, i.e., in the red and blue light regions. This was the first demonstration of an action spectrum. In brown and red algae, the presence of accessory pigments filled the “green gap” where photosynthesis also occurred and bacteria would accumulate.

Thus, his experiments showed that green algae photosynthesised best in red and blue light wavelengths, matching the absorption spectrum of chlorophyll-*a*. Brown algae performed better in yellow-green wavelengths, and red algae favoured green and blue light, corresponding to their pigments.

Bringing all of this back to Ørsted's 1844 predictions, Engelmann's results appeared to confirm that green algae thrive in shallow waters with abundant red and blue light; brown algae are found in intermediate depths with greenish light; and red algae in deep waters with blue light. While this seemed to offer a mechanistic explanation for the distribution of algae colours in relation to ocean depth, it was mostly theoretical and experimental at that time; actual field confirmation came later.

Even so, Engelmann's work was foundational and influenced understanding up until recently; many zoologists taught these findings as fact. Later work by researchers like Haxo and Blinks added supporting evidence, but eventually, new perspectives emerged, something we'll cover in upcoming lectures.

2.1.b Part II

We're going to continue with our story on chromatic adaptation and look at some of the work Haxo and Blinks did, probably about 50 or 60 years after Engelmann's work. They repeated experiments similar to what Engelmann did, but with newer and more modern technology. They used devices developed in their laboratories that could precisely create monochromatic spectra of light. They used a variety of red, brown, and green seaweeds, as Engelmann did. However, they also examined the absorption spectrum of light by various intact thalli of the seaweeds, as well as extracts of pigments obtained from the different seaweeds. Thus, they had three lines of evidence used in combination. Engelmann only looked at action spectra, but Haxo and Blinks combined action spectra with absorption spectra determined in thalli as well as in extracts. Based on all of this accumulating evidence, they made various conclusions.

When they did these experiments on green algae, they showed that the thallus absorbs light best in the blue light (around 435 nm) and red light (675 nm) regions. The thallus absorption matched very closely the absorption spectrum obtained from a chlorophyll-*a* extract. They also found that the absorption spectrum in the thallus closely matched the action spectrum determined for green algae. In Engelmann's experiments, the accumulation of aerotactic bacteria was used, but Haxo and Blinks actually measured the rate of photosynthesis. In their graph for green algae, absorption peaks for chlorophyll-*a* are in the blue and red light regions. The solid line with open dots represents the absorption spectrum, indicating how the thallus absorbs light across tested wavelengths, while the dotted line is the action spectrum, the amount of photosynthesis at each wavelength.

At the peak chlorophyll-*a* absorbance in the blue and red light regions, the action spectrum closely matches the absorption spectrum. In the region from around 460 to 500 nm, there's a major discrepancy. In this region, carotenoids such as beta-carotene can capture light not accessible to chlorophyll-*a*, then pass it on to chlorophyll-*a* to drive photosynthesis at additional wavelengths. This region shows the effect carotenoids have on enhancing photosynthesis where chlorophyll-*a* alone could not. This clearly demonstrates the role of accessory pigments in harvesting light not typically accessible to chlorophyll-*a*, passing it on, and resulting in photosynthesis in that region.

For brown algae, the accessory pigment is mainly the xanthophyll fucoxanthin, which gives those seaweeds their colour. Fucoxanthin improves the algae's ability to absorb light in the green to

yellowish regions (500–560 nm). The presence of chlorophyll-*c* also helps with absorption around 630 nm. Some species contain so much chlorophyll-*c* that they appear almost optically black, i.e., opaque to all light. Again, chlorophyll-*a* absorbs maximally in the blue and red light, but in the region that would normally be the green gap, the presence of xanthophylls allows for absorption of light and passage to chlorophyll-*a*, thereby enabling photosynthesis that would not occur without xanthophylls. Xanthophylls, specifically fucoxanthin, capture light in the green gap region (and assisted by some chlorophyll-*c*), passing it on to chlorophyll-*a* and enabling more effective photosynthesis in green and yellowish, bluish-yellowish light.

Red seaweeds show similar patterns where the absorption and action spectra match closely, except for an anomaly around 500–570 nm. Explanation for this came later; it's called the Emerson enhancement effect, a complex photophysiological response seen primarily in red algae. If you're interested, you can read the seminal paper by Rajni Govindjee and Govindjee from 1964, which is still widely cited in photosynthesis research.

Overall, Haxo and Blinks' work provides further confirmation of Emerson's work from about 60 or 70 years earlier. Mary Beth Saffo's review concludes that Haxo and Blinks' work was generally interpreted as a modern confirmation of Engelmann's work. Haxo and Blinks' conclusions were a bit more cautious regarding statements about the vertical distribution of seaweeds. They acknowledged, specifically regarding red algae with phycobilins, that it seems logical to assume the vertical distribution of red algae is influenced, at least in part, by these pigments. Some species may be unable to extend their distribution to depths not readily penetrated by other algae. Thus, they cautiously concluded that red algae, by virtue of red pigments absorbing blue light, can extend further down in the water column, attributing this vertical extension at least in part to the presence of phycobilins. However, they also noted that other algae can also be found at depth, and red algae are found in shallow waters as well. Real-life observations don't always match the expectations from Haxo, Blinks, and Engelmann's work. Nonetheless, there is some support for red algae appearing deeper down.

They also emphasised that algal distribution is influenced by many factors, all of which must be considered. These factors will be addressed later in the third part of the chromatic adaptation lectures.

Are Engelmann's conclusions valid? Over the years, especially more recently, with greater access to deep algae and more sampling, evidence has mounted to suggest that the widely accepted hypothesis (green algae at shallow depths, red at deep depths, brown at intermediate depths) is not well-supported when looking at actual distributions. As you can see from Saffo's paper, there is significant overlap: green, brown, and red algae share similar ranges in both minimum and maximum depths, though red algae may reach as far as 160 meters deep. However, a discovery 15–20 years ago showed a green algae species holds the record for deepest living seaweed. So empirical evidence no longer matches the theoretical expectations of Engelmann's hypothesis: the colour or adaptive ability inferred by pigments is not a reliable indicator of the environment in which the seaweed is found. Thus, the theory of chromatic adaptation doesn't seem to hold up in practice.

What really explains vertical distribution in the ocean? To answer this, we'll have to look at work by Ramus, Rosenberg, and Ramus, which will be the topic of our next lecture.

2.1.c Part III

Right, so now we're going to look at the third part of our series of lectures on chromatic adaptation. This time, we're going to look at some *in-situ* experiments that examine how seaweeds adapt and become acclimatised to different light regimes within the ocean. This is a very different approach compared to the previous studies by Engelmann, Haxo, and Blinks, which were based on laboratory studies looking at seaweeds in isolation rather than how they respond adaptively to environmental changes. Rosenberg and Ramus' work is quite different in this regard. We're going to specifically look at work by John Ramus – experiments he conducted in the late 1970s. These were innovative, “funky” experiments focusing on acclimatisation as a process by which seaweeds, regardless of colour, adapt to different light regimes in the ocean.

They took seaweeds that had been acclimatised (meaning they'd been grown at shallow depths near the surface for a couple of weeks, allowing their photo-physiological equipment and pigment composition to adapt to those environments) and compared them to seaweeds grown simultaneously at deeper depths (about 10 meters), which had also become acclimatised to those conditions. Given that seaweeds come in different colours, they used red and green seaweeds for these experiments. Remember, the red algae have phycobilins as well as some beta-carotene as accessory pigments, and the green algae mostly have chlorophyll-*a* and some beta-carotene, with definitely no phycobilins in the green algae.

They used four species: *Chondrus crispus* (red), *Porphyra umbilicalis* (red), *Codium fragile* (green), and *Ulva lactuca* (green). These also represent two functional forms: coarsely branched varieties (*Chondrus* and *Codium*) and membranous varieties (*Porphyra* and *Ulva*). By using different seaweed colours and classifications within the functional form model, they designed transplant experiments to figure out how algae become adapted to different light regimes in the ocean.

These slides show that they're comparing within the same functional form group: *Ulva* and *Porphyra* (green and red), both acclimatised to live at a depth of about 10 meters for a couple of weeks. When you compare their photosynthetic rates per unit of chlorophyll-*a*, *Ulva lactuca* and *Porphyra umbilicalis* have exactly the same rate of photosynthesis. Chlorophyll-*a*, whether in *Ulva* or *Porphyra*, drives the same amount of photosynthesis in the two species. However, when expressing photosynthetic rate per gram of tissue, *Ulva*'s rate is higher than *Porphyra*'s, because a gram of *Ulva* contains more chlorophyll-*a* than a gram of *Porphyra*. This is because chlorophyll-*a* is the predominant pigment in *Ulva*, while *Porphyra* relies more on accessory pigments like phycobilins.

They also compared red and green species at 10 meters, focusing on differences in surface area to volume ratios. Species with high surface area to volume ratios exhibit higher rates of photosynthesis per gram of tissue, which is predictable based on the functional form model for seaweeds.

Further experiments involved transplanting specimens acclimatised to 10 meters to the surface (0 meters), where light is more abundant and of different quality, with more red wavelengths present. After transplantation, the photosynthetic rate per gram of tissue at the surface doubles

compared to the same species still at 10 meters. The reverse experiment (moving surface acclimated specimens to 10 meters) results in a drop in photosynthetic rate, due to decreased light availability.

When comparing pigment content between surface-acclimated and deep-acclimated specimens, the latter have around ten times higher chlorophyll-*a* concentrations, as well as a higher chlorophyll-*b* to chlorophyll-*a* ratio (chlorophyll-*b* being an accessory pigment). This ratio increases in seaweeds adapted for dimmer deep water.

Codium (the coarsely branched green alga), on the other hand, shows no significant difference in photosynthetic rate per gram of tissue regardless of being acclimated at shallow or deep depths. Comparing pigment concentrations, deep-acclimated *Codium* has only 1.5 times more chlorophyll-*a* and -*b* compared to surface-acclimated specimens, and the ratio of chlorophyll-*b* to chlorophyll-*a*, or photosynthetic rate per gram of chlorophyll-*a* or per unit surface area, shows no difference between shallow and deep specimens.

Another comparison shows that after acclimation to low-light environments, the photosynthetic rate per gram of chlorophyll-*a*, and per unit of surface area, is higher in *Codium* than in *Ulva*.

These findings suggest that seaweeds exhibit a high degree of physiological plasticity, changing pigment concentrations and photosynthetic responses as they acclimatize to new light conditions, though *Codium* is less plastic in its acclimation response compared to *Ulva*.

For *Ulva*, after an instantaneous transplant from the surface to 10 meters, photosynthetic rate drops due to lower light. Over weeks, the seaweed responds by manufacturing more chlorophyll-*a* and -*b*, increasing light-harvesting ability, so that the rate of photosynthesis at depth improves significantly. The chlorophyll-*b* to chlorophyll-*a* ratio also increases under low light, boosting the light-harvesting ability when light is limiting. The reverse, moving *Ulva* from deep to shallow, results in an immediate rise in photosynthesis, while pigment concentrations decrease over time. However, per unit of chlorophyll-*a*, the photosynthetic rate remains constant, reflecting the unchanging role of chlorophyll-*a* in photosynthesis.

Interpretation of *Codium* results shows a very different response. Despite sharing the same pigments as *Ulva*, *Codium* doesn't show the same changes when transplanted between different light levels. *Codium*'s internal anatomy, with its thallus thickness (about 3 mm) and "coenocytic" construction (no cell walls, multinucleate, woven filaments), enables it to pack an abundance of chlorophyll-*a* around the periphery of its utricles. The result is a surface that is optically black – nearly all incident light is absorbed, making *Codium* highly effective at light capture regardless of light conditions. This explains *Codium*'s limited response in transplant experiments and its relative lack of plasticity compared to *Ulva*.

Ulva, by contrast, is very thin (about 17 microns), like green cellophane. All cells in its high surface area to volume ratio structure are exposed to light, but light is not all absorbed, as there is always more than enough. In deeper water, *Ulva* ramps up pigment production to absorb a higher proportion of available light. However, due to the lack of certain accessory pigments, there is always some light in the "green gap" region that *Ulva* cannot use.

Meanwhile, *Codium*, despite lacking these accessory pigments, can still absorb light across the green gap thanks to its optically black surface and dense arrangement of pigments, making it effective at light capture in both shallow and deep waters. Its specific internal arrangement (thick filaments, syncytic thallus, and peripheral chlorophyll-*a*) explains why *Codium* behaves differently from *Ulva* and why it is less plastic in its acclimation responses.

These experiments reveal key differences in how seaweeds adapt to varying light environments, with *Ulva* exhibiting high plasticity in pigment composition and photosynthetic capacity, while *Codium* achieves efficient light capture through a different anatomical and physiological strategy, showing less change across light environments. This concludes our discussion on light capture and chromatic adaptation in these seaweed species.

Bibliography