

Subject: Scholes cabin 5: Phytoplankton Fluorescence

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Dear Stirling,

There are another two instruments in my lab that I haven't described yet. They are both fluorometers, but of different sorts. Fluorometers measure the glow that plants give off when you shine a bright light on them. The reason they do so is because you overload their photosynthesis system, and they have to get rid of the extra energy to protect the delicate machinery. To understand how these instruments work I need to describe how photosynthesis works in some detail. Hold on tight, this is university-level stuff! But I know how you love complicated machinery. If you get stuck, ask Mom, she is a photosynthesis expert.

I don't need to remind you what an amazing 'invention' photosynthesis was. It transformed our planet, 3.5 billion years ago, into the green, oxygen-rich place that we love. It is so complicated and so effective that the basic elements only seem to have evolved once, and are shared by all photosynthetic organisms from bacteria to boobies. The green stuff in leaves (and phytoplankton) is a pigment called chlorophyll.

Actually, several different pigments help out in the story I am going to tell you, especially in plankton, but I am trying to keep it simple! A photon of light excites a molecule of chlorophyll which is part of a tiny machine called 'photosystem 2'. From then on out it is a game of pass-the-parcel: each step unloads the energy onto the next step, and until it does so it can't accept any more. If the whole chain is blocked, then the chlorophyll has no choice but to dump the energy as a fluorescent glow. Step one is to pass the energy from the 'antenna' where the photon is captured to the 'Reaction Centre 2' which quickly oxidises (in other words, steals an electron from) a region of the chlorophyll molecule called P680. P680 then passes the energy, now in the form of an electron, on to a quinone (called Qa), which unloads it on another quinone, Qb. When Qb has collected two electrons, it can finally be freed from the reaction centre, and rest for a moment as part of the 'plastoquinone pool', a sort of a battery. From there, helped by another pigment called cytochrome, the electron is passed on to 'photosystem 1', where more light energy is captured. The energy is finally used by the Calvin cycle to bolt CO₂ molecules together into sugars. Now the pipeline is clear for another photon to be captured!

But I left out an important bit: what happens to the oxidised P680? It gets a replacement electron ultimately from the breaking up of a water molecule through another chain of steps, which liberate O₂ in the process (Yay! We need that!) and H⁺, which becomes part of the sugar hydrocarbons we are building. Phew! What a complicated machine,

and all happening in about 1/20th of a second in a molecule too small to see, even with a microscope.

But we can tell what is going on by putting way too many photons in at the beginning, and then seeing where the energy parcel gets stuck. The continuous flow fluorometer just zaps a tube of flowing seawater with bright blue light and then measures the fluorescent glow in a specific wavelength (680 nm). That tells us how much chlorophyll is in the sample. Here, near the ice, the numbers are much higher than they were in the open ocean.

The other instrument is a fast-repetition fluorometer, also called Fluorescence Induction and Relaxation, or FIRe. It blasts with a bright but short pulse of light, enough to send a single wave of electrons down the cascade, and watches how quickly the photosystem 2 fills up and starts shedding energy through fluorescence. If PS2 unloads almost everything right away, it means the whole machine is switched off. So, although chlorophyll may be present, the phytoplankton is prevented from doing photosynthesis by something else - often a shortage of nutrients. The instrument then gives a long blast of light and watches how the fluorescence dies down when the light is switched off. That tells us just where on the chain the electrons are getting stuck.

It turns out that I know the man who developed this clever instrument, Paul Falkowski. Science is really quite a small world. Paul jokes that all the really important evolutionarily inventions - like photosynthesis - were made quite early on. Humans are just bacteria with a brain!

Love,

Dad