

Interactive comment on “Interactive effects of and light on growth rates and RUBISCO content of small and large centric diatoms” by G. Li and D. A. Campbell

Anonymous Referee #4

Received and published: 2 December 2015

General Comments: This paper examines the effects of light and nitrogen availability on cellular growth rates, allocation of N to different functional pools and photo-physiology in a large and small diatom. The authors use carefully conducted turbido-stat culturing methods, and a variety of molecular / physiological assays to document somewhat different responses to light and nitrogen levels in the two species. A variety of results are reported, and I found it difficult to extract one main message from the paper. Some of the more salient results include: differential apparent growth response to N in the two species (larger diatom shows lower growth in high N medium); 2) increased allocation of N to RubisCO and PSII in low N and/or low light growth conditions. Without better quantification of NO₃ levels in the 'high nitrogen' experiments, I find it difficult to

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interpret the results. I am not an expert on N uptake by phytoplankton, but I'd expect half saturation constants for growth (K_m) should be on the order of 2 - 5 μM NO₃. So I think it is very important to have better data on the N concentrations in the medium (i.e. more than simply ' $<10 \mu\text{M}$ '). My other general comment concerns the integration of various data sets. A significant amount of FRRF data is presented, but I don't feel that these have been utilized to their potential to provide mechanistic understanding of the results. For example, these data are not even mentioned in the discussion.

Specific Comments: I concur with reviewer 1's assessment that the paper is difficult to read. I had to go over the results section many times, flipping back and forth between the figures to try extracting some main ideas. This may be in part because the patterns are not necessarily clear and no overall generalizations can be drawn. That is ok, in principal, and the authors can't be faulted for this. However, I feel that the writing was unnecessarily complex in places and could be improved to enhance clarity. For example, the abstract says:

Low nitrogen decreased the growth rate of the smaller diatom, particularly under higher light, but stimulated the growth rate of the larger diatom. Our results show that the high nitrogen in common growth media favours the growth rate of a small diatom but inhibits growth of a larger species .

There is a subtle, but potentially significant, change in language that makes the writing more complex than necessary. You could argue that low N inhibition or high N stimulation are the same thing, but it would be best to focus on one side of this coin.

With respect to FRRF measurements, the 5 min time-scale is likely too short for proper 'dark-adaptation'. I think people have begun using the term 'dark-regulated' instead.

Top of p.16654 growth rate 'significantly higher for the larger diatom in LN medium. Stats? The scaling on Fig. 1b makes it hard to see the changes. Table 1: p. 16654, bottom. 'weak downward trend with increasing light'. What is the reader supposed to take from this (and many other 'wooly' sentences)? Is this a statistically significant

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result? Otherwise, it should be reported as no change.

Same paragraph: From table 1, I see no clear change in prot:cell across the N treatments. At 30 and 90 light levels, protein is lower under low light. This is the opposite of what is stated in the results.

Same page ~ line 21. What is the significant of a change from 1085 to 896 pg cell⁻¹. Is this ~10% change statistically significant, and mechanistically important?

Here is an example of a sentence that is (in my opinion) overly wordy and thus not effective in driving home a main point:

The molar fraction of N allocated to RUBISCO relative to total cellular N under high nitrogen slightly increased from 0.061 to 0.086 for *T. pseudonana* as growth light increased and varied from 0.019 to 0.034 with no clear trends for *T. punctigera*. (Fig. 2g and h).

It seems to me that that there are quite a few different ideas packed into this sentence. Which are the main ones?

p. 16655 line ~ 15. Fv/Fm 'ranged' from 0.57 to 0.59? Isn't this virtually the same number? I'd say it was essentially invariant.

Sampe page. Line ~ 19 I'd say 'down-regulation' of PSII rather than 'inhibition'

p. 16656 line ~ 5. Does high N medium 'inhibit' the rate of e transport, or does low N growth increase this rate. Again, this is subtle different in language, but mechanistically and conceptually, I think it's important.

First paragraph of discussion. Fig. 2 shows an apparent decrease in N allocation to RubisCO under N-rich media. The text says the opposite.

In general, I found that the discussion didn't really provide much mechanistic insight into the results, and I didn't come away with a clear message from the paper.

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Interactive comment on Biogeosciences Discuss., 12, 16645, 2015.

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