Identifying *Dunaliella*'s Iron-Starvation Responses for Photosynthetic Maintenance

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ABSTRACT

Iron is often a limiting nutrient in nature, with 30% of the world's oceans and 40% of arable land containing so little bioavailable iron that it limits primary productivity (Moore and Braucher 2007). Photosynthesis is an iron heavy process, so in low iron environments, photosynthetic efficiency is reduced in many organisms. The eukaryotic, green algae *Dunaliella* are extremophiles that are found in saline environments and have been shown to grow in environments with extremely low iron. In this paper I aimed to identify the mechanisms iron-starved *Dunaliella* employs to maintain high levels of photosynthetic efficiency. To do this, I first determined the pre-defined iron nutritional stages of iron-limited, deficient, and replete conditions for two species of *Dunaliella*: *Dunaliella tertiolecta* and the beta-carotene hyper-accumulating *Dunaliella salina.* I discovered that the iron limited and replete conditions for both species of *Dunaliella* are 0.15 and 15 uM Fe respectively. The iron deficient condition for *D. salina* is 3 uM Fe and for *D. tertiolecta* it is 5 uM Fe. I then compared the photosynthetic efficiency parameters of both *Dunaliella* species to the reference alga, *Chlamydomonas reinhardtii,* across all three iron conditions. *D. salina* seems to maintain its photosystem II (PSII) efficiency better than *D. tertiolecta* under low iron nutrition. I hypothesize that this may be due to its slower growth rate or a better low iron uptake ability. Understanding how the unique low iron mechanisms function in *Dunaliella* will give insight into engineering other photosynthetic organisms to respond better to low iron nutrition, which could lead to an increase in the world's crop yields, primary productivity, and carbon sequestration.

KEY WORDS

Iron homeostasis, *Chalmydomonas reinhardtii, Dunaliella salina, Dunaliella tertiolecta*, photosynthetic efficiency

INTRODUCTION

Iron is an abundant element in nature. However, its Fe(II) oxidation state, which is the form organisms utilize, is often scarce. This is because when iron reacts with oxygen, it forms insoluble Fe(III) oxides. Since we live in an aerobic planet, this results in 30% of the world's oceans and 40% of arable land being considered iron-limited (Moore and Braucher 2007). Photosynthesis is a major iron sink, requiring 30 atoms of iron, assuming a one-to-one stoichiometry of the major photosynthetic complexes (Blaby-Haas and Merchant 2017). When iron becomes limiting and can no longer meet the demands of photosynthesis, iron-limitation typically results in a reduction in photosynthetic efficiency which leads to reduced crop yields (Zuo and Zhang 2009) and carbon sequestration (Blain et al. 2007). Therefore, it is important to research strategies to combat the effects of iron-limitation.

The response of algae and other photosynthetic organisms to low iron nutrition depends on environmental iron availability. A common phenotype of iron-limitation in photosynthetic organisms is chlorosis, the reduction of chlorophyll that results in the yellowing of plants and algae. The Merchant Lab has defined three iron nutritional stages in green algae to study responses to iron nutrition: iron-replete, iron-deficient, and iron-limitation (Glaesener, Merchant, and Blaby-Haas 2013). "Iron-replete" is the iron concentration at which algae does not display chlorosis and does not activate iron starvation responses such as the induction of low iron transporters to increase the iron imported into the cell "Iron-deficient" is the iron concentration at which algae do not show signs of chlorosis but start to activate iron starvation responses. "Iron-limited" is the iron concentration at which algae show signs of both chlorosis and iron starvation responses. This reduction of chlorophyll ultimately reduces the efficiency of photosynthesis (Blaby-Haas and Merchant 2017). One of the negative consequences associated with a reduction in photosynthetic efficiency is the reduction of agricultural crop yield, which is one of the contributing factors for global hunger (Watson et al. 2000).

Photosynthetic organisms developed different mechanisms for coping with low iron conditions. For example, iron-stressed cyanobacteria will induce an iron-replacing mechanism by upregulating the iron-stressed induced (*isi)* operon. (Chauhan et al. 2011, Laudenbach and Straus 1988). The *isi* operon encodes the iron-replacing protein flavodoxin, which is a non-iron, flavin-containing cofactor that reduces the iron requirement of photosynthesis by functionally replacing

ferredoxin, an iron-sulfur cofactor which is used to transport electrons from photosystem I to reduce NADP+ to NADPH. In addition, the *isi* operon encodes the isiA protein, which attaches to photosystem I to help enhance light capture. In many organisms, photosystem I is a target for degradation when iron becomes limiting due to its high iron content at 12 iron atoms. The well-studied freshwater eukaryotic, green alga *Chlamydomonas reinhardtii* can be grown solely on the carbon it produces from photosynthesis, but also assimilate carbon from the media in the form of a reduced carbon source, acetate. In iron deficiency, *C. reinhardtii* degrades photosystem I and LHC1 antennae under low iron nutrition when grown photoheterotrophically (Moseley et al. 2002), but when grown photoautophically, *C. reinhardtii* maintains its photosynthetic efficiency (Terauchi et al. 2010). It was hypothesized that photoautophically grown *C. reinhardtii* maintains the photosynthetic apparatus since it depends on photosynthesis for its sole carbon source to grow and develop. There are a wide variety of mechanisms iron-starved, photosynthetic organisms have evolved. Therefore, before we engineer organisms to thrive in low iron environments, we must identify the variety of mechanisms used to maintain photosynthetic efficiency during iron starvation.

Dunaliella tertiolecta and *Dunaliella salina* are halotolerant eukaryotic, green algae. They are known to respond particularly well to low iron nutrition, capable of even surviving on media with no additional iron, however *Dunaliella's* maintenance of photosynthetic efficiency in low iron nutrition has not been systematically studied. Iron-stressed *Dunaliella* are capable of functionally replacing ferredoxin with flavodoxin (Blaby-Haas and Merchant 2017) and also induces a novel chlorophyll a/b binding protein, Tidi, which attaches to Photosystem I to enhance light capture (Varsano et al. 2005). A key difference between *D. tertiolecta* and *D. salina* is that *D. salina* hyper-accumulates **β**-carotene while *D. tertiolecta* does not. **β**-carotene is a carotenoid that helps dissipating harmful, excess light as heat and can also act as an antioxidant, which reacts with reactive oxygen species (ROS) to eliminate their free radicals (Paiva and Russell 1999). Iron limited cells become especially susceptible to ROS, since there is less production of chlorophyll and photosynthetic complexes which bind chlorophyll are degraded (Moseley et al. 2002), so *D. salina*'s ability to accumulate **β**-carotene could be an important mechanism help to prevent ROS damage to chloroplast DNA, proteins, and photosynthetic apparatus under low iron nutrition. By comparing two species of *Dunaliella,* it may be possible to identify the mechanisms to maintain

photosynthetic efficiency that are independent to the induction of Tidi or flavodoxin, such as the accumulation of **β**-carotene, differences in growth rate, or differences in cell size.

I seek to understand the diversity of how photosynthetic efficiency is maintained in divergent environments by comparing *Dunaliella* against other species of algae. Through these series of experiments, I aim to answer the question "what mechanisms are allowing Dunaliella to maintain their photosynthetic efficiencies under low iron nutrition?" I will answer this question in two phases; (1) identifying *Dunaliella'*s iron-nutrition needs that corresponds to pre-defined iron-replete, -limited, and -deficient nutrient phases and (2) measuring photosynthetic efficiency across the different iron conditions and comparing the two Dunaliella species against the reference alga, *C. reinhardtii*. I hypothesize that the photosynthetic efficiency of *Dunaliella* in low iron nutrition is maintained because the bioenergetic exchange of ferredoxin for flavodoxin reduces the total number of iron atoms necessary for photosynthesis and the induction of Tidi to compensate for reduced light harvesting because of the reduction of Photosystem I content. Understanding these mechanisms will give insight into engineering other photosynthetic organisms to respond better to low iron nutrition, with the potential to increase global primary productivity, agricultural crop yields, and oceanic carbon assimilation (Watson 2000, Falkowski 1998).

METHODS

Strains and Culture Conditions

I used *D. salina* LB 2538, *D. tertiolecta* LB 999, and *C. reinhardtii* CC 4533 for the following experiments. Medium was prepared in acid washed glassware to remove contaminating iron. I grew *D. salina* and *D. tertiolecta* in media described by (Pick, Karni, and Avron 1986) with varying concentrations of Fe added from a solution of $FeCl_3$. I ran the NaCl for the Polle Pick media through a chelax column to remove trace metal contamination. I grew *C. reinhardtii* in acetate-free Tris-Phosphate media with varying concentrations of Fe added from a solution of Fe chelated with EDTA. I grew all the liquid cultures at 24ºC with 140 r.p.m. shaking and a light intensity of 60 $m^{-2}s^{-1}$. I inoculated cells of *D. salina* at a concentration of 2 x 10^5 cells/mL, and I inoculated cells of *D. tertiolecta* and *C. reinhardtii* at a concentration of 1 x 10^4 cells/mL.

Protein isolation for immunoblot analysis

To detect the presence and abundance of specific proteins, I used a combination of 2D gel electrophoresis to separate proteins by molecular weight and then immunodetection to stain specific proteins. To collect *Dunaliella* cells, I centrifuged 40 mL of cultures at 1800 rpm for 3 minutes. I discarded the supernatant and resuspended the cells in a sodium phosphate buffer. To ensure equal amounts of protein in each sample that I loaded into the electrophoresis gel, I conducted a BCA protein assay and used a Nanodrop UV-Vis Spectrophotometer to measure the proteins content in each sample. Then I diluted the samples with SDS to 1 μg/mL and placed the samples in a 65ºC water bath for 20 minutes to ensure that they were fully denatured. I then loaded 10 μg of protein from each sample into a 20% polyacrylamide gel and transferred onto a 0.1 uM nitrocellulose membrane for 1 hour under a constant current of 60 mA. Membranes were blocked for 1 hour in 3% (w/v) dry milk in PBS with 0.05% (w/v) Tween 20. Membranes were incubated in primary antibodies diluted in PBS with 0.05% Tween 20 for 1 hour. Primary antibody dilutions were at 1:5000 for CF_1 and 1:500 for FLV. Membranes were then washed in PBS with 0.05% Tween 20 briefly twice and then for 15 minutes and then 3 times for 5 minutes. The secondary antibody was alkaline phosphatase conjugated goat anti-rabbit IgG used at 1:6000. Proteins were visualized using the alkaline phosphatase color reaction described by Sambrook and Russell (2001).

Chlorophyll Determination

To determine chlorophyll content, I aliquoted 1 mL of culture into 1.5 mL Eppendorf tubes and centrifuged them for 1.5 min at 1500 rpm at 25 ºC in a microcentrifuge. I removed the supernatant so that only the cells remained in the tubes. To extract the cells' chlorophyll, I added 1 mL of a solution consisting of 20% methanol and 80% acetone to the tubes. To remove the cell debris and non-soluble proteins, I centrifuged the tubes at 13,200 rpm for 2 minutes at room temperature. I measured the chlorophyll content of each sample using a UV-6300PC spectrophotometer to measure their absorbance of 750, 663.6, and 646.6 nm wavelength light and normalized by cell density to obtain units of chlorophyll/cell (Porra et al. 1989). Statistical

significance for chlorophyll content reduction was calculated using a t-test and the significance threshold was $p < 0.05$.

Determining Iron Levels

To study the algae's responses to low iron nutrition, I first determined the iron replete, deficient, and limited conditions for *D. salina* and *D. tertiolecta.* To do this, I grew cells of *D. salina* and *D. tertiolecta* in media containing varying amounts of iron from 0 – 150 μM Fe. Each iron concentration had three replicates. Once cultures reached mid-log phase (3×10^6 cells/mL for *D. tertiolecta* and 1×10^6 cells/mL for *D. salina*), I measured their chlorophyll content. I used a t-test to determine which iron concentrations had a significant reduction in chlorophyll content compared to the iron replete condition. I detected the induction of flavodoxin with immunoblot analysis as a low iron stress marker (La Roche, 1993).

Comparing Photosynthetic Efficiency

To assess the photosynthetic efficiency of *Dunaliella'*s low iron response, I compared the low iron responses of *D. salina, D. tertiolecta*, and photoautotrophically grown *C. reinhardtii* in their respective iron-replete, deficient, and limited conditions. Once cultures reached mig-log phase, I collected cells for immunoblot analysis to identify the increased abundance of iron status markers such as FOX1 in *C. reinardtii* and FLV in *D. salina* and D. *tertiolecta* and I measured the chlorophyll per cell to ensure that the cells were iron limited, deficient, and replete. To test each cells' photosynthetic efficiency, I measured F_v/F_m, which measures Photosystem II efficiency. To do this, I aliquoted 300 μL of culture onto 96 well plates, dark acclimated the cells for 10 minutes, and then measured the F_v/F_m using an Imaging PAM. I then used t-tests to determine the significant differences in the chlorophyll content and F_v/F_m for each algal species across their iron replete, deficient, and limited conditions.

RESULTS

Determining Iron Levels

In order to determine the iron replete, deficient, and limited conditions for *D. salina* and *D. tertiolecta*, I first grew the two species in a wide range of iron conditions consisting of 0, 0.15, 1.5, 15, and 150 μM Fe to isolate the range at which cells become low iron stressed. I measured how the chlorophyll per cell and the abundance of flavodoxin changed across these conditions. I found that as the iron concentration increases, so does the chlorophyll per cell in both species of Dunaliella. This increase levels off at 15 μM Fe and there is no significant increase in chlorophyll in the 150 μM Fe condition. In the 15 μM Fe condition, there is also no induction of flavodoxin, so therefore I selected 15 μM Fe as the iron replete condition for both species of *Dunaliella*. Since the first criteria for the iron deficient and limited conditions depend on their chlorophyll contents relative to the replete condition, I used a t-test to identify which iron conditions had a significant reduction in chlorophyll compared to the 15 μM Fe condition. I found that there is a significant reduction in chlorophyll in *D. salina* in the 0 and 0.15 μM condition (Figure 1A) and in *D. tertiolecta* in the 0, 0.15, and 1.5 μM condition (Figure 1B). While not statistically significant, there is also a large visual reduction in the chlorophyll content of *D. salina* grown in 1.5 μM Fe. Since the second criteria for the iron deficient and limited conditions depend on the induction of low iron stress markers such as flavodoxin, I performed an immunoblot to test for the presence of flavodoxin each of my iron conditions. I found that flavodoxin is present in both algal species in the 0-1.5 μM Fe conditions (Figure 1). Therefore, I determined that my iron limited condition for both species of *Dunaliella* were 0.15 μM Fe. Another indicator of iron limitation is a reduction in growth. Therefore, to verify that 0.15 μM Fe is the iron limited condition, I followed the growth of *D. salina* and *D. tertiolecta* across all 5 iron conditions. I found that there is a reduction in growth of *D. tertiolecta* in both high iron (150 μM Fe) and low iron (0 and 0.15 μM Fe) conditions compared to 15 μM Fe (Figure 2B). However, surprisingly in D. salina, there is no significant difference in growth between iron conditions (Figure 2A). These results are enough to conclude that 0.15 and 15 μM Fe are the iron limited and replete conditions for *D. salina* and *D. tertiolecta*. However, since there was a reduction in chlorophyll in both *Dunaliella's* 1.5 μM Fe condition, I

repeated this experiment with new iron conditions consisting of 1.5, 2, 3, 4, 5, and 15 µM Fe to determine *D. salina* and *D. tertiolecta's* iron deficient conditions.

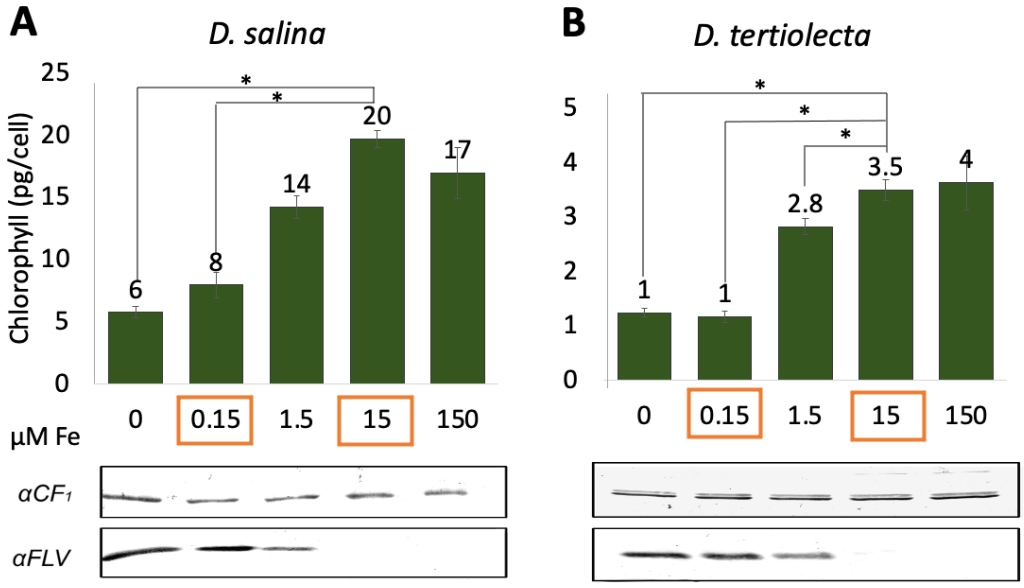

Figure 1. Chlorophyll content and immunodetection of flavodoxin for (A) *D. salina* and (B) *D. tertiolecta* under varying iron conditions. CF_1 is included as a loading control. Iron limited and replete conditions are boxed in orange. Significance is based on a t-test (p-value ≤ 0.05).

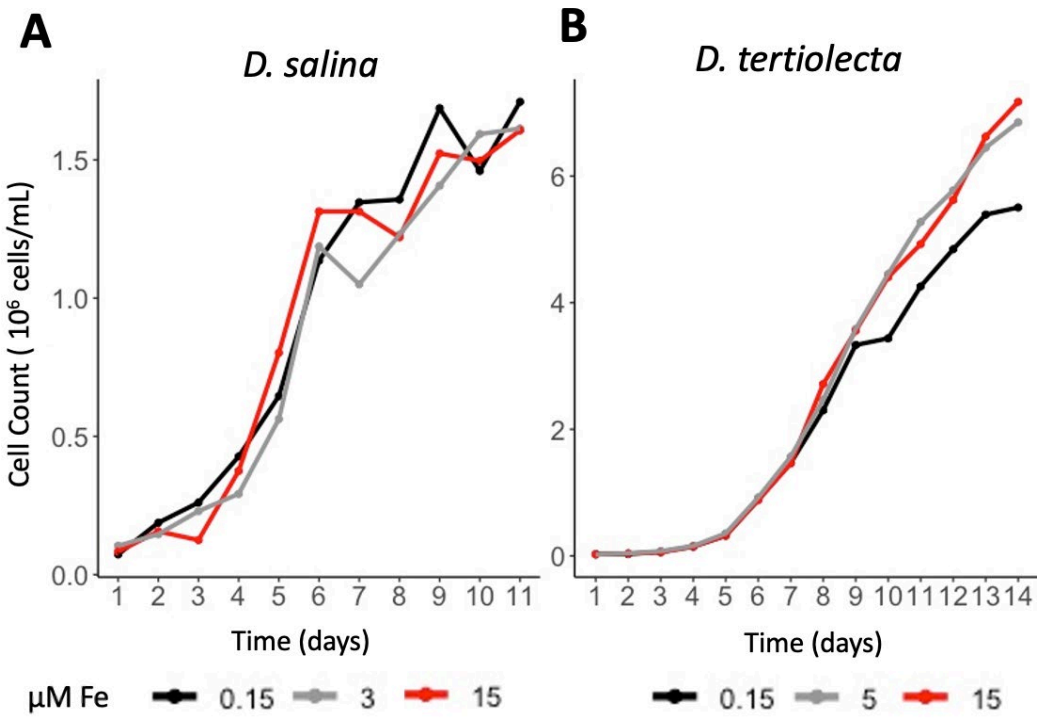

Figure 2. Growth curves of (A) *D. salina* and (B) *D. tertiolecta* under iron limited, deficient, and replete conditions.

In this new round of iron conditions ranging from 1.5-15 µM Fe, I found that there is no significant reduction in chlorophyll in any of the conditions when compared to the iron replete 15 µM Fe condition and that flavodoxin is induced from the 1.5-3 µM Fe conditions in *D. salina* (Figure 3). While I could have selected 1.5, 2, or 3 µM Fe to be the iron deficient condition for *D. salina*, since each of those conditions contained flavodoxin and did not have a significant reduction in chlorophyll, I decided to choose 3 µM Fe for *D. salina*'s iron deficient condition because its chlorophyll content was the closest to the replete condition.

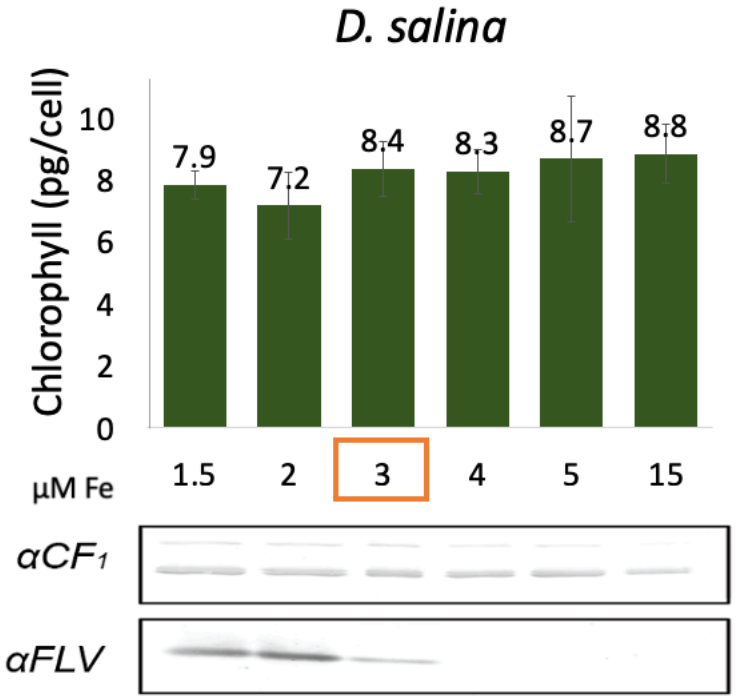

Figure 3. Chlorophyll content and immunoblots of FLV for *D. salina* under varying iron conditions. The iron deficient condition is boxed in orange. Significance was calculated using a t-test (p-value ≤ 0.05).

When I completed the same experiment for *D. tertiolecta*, I was unable to identify its iron deficient condition. I found that flavodoxin is induced in all of the 1.5-5 µM Fe conditions and there is no significant reduction in chlorophyll in any of the 1.5-5 µM Fe conditions when compared to the iron replete 15 µM Fe condition (Figure 4A). Therefore, to find out if there was an even higher the concentration at which flavodoxin is expressed, I repeated my experiment again with a higher range of iron concentrations consisting of 5, 6, 7, 8, 9, and 15 µM Fe. I found that the 5 µM Fe condition is the highest concentration where flavodoxin is expressed (Figure 4B).

Since out of all the iron concentrations where flavodoxin is expressed 5 µM Fe has the chlorophyll content closest to that of the replete condition, I selected 5 µM Fe to be the deficient condition for *D. tertiolecta.*

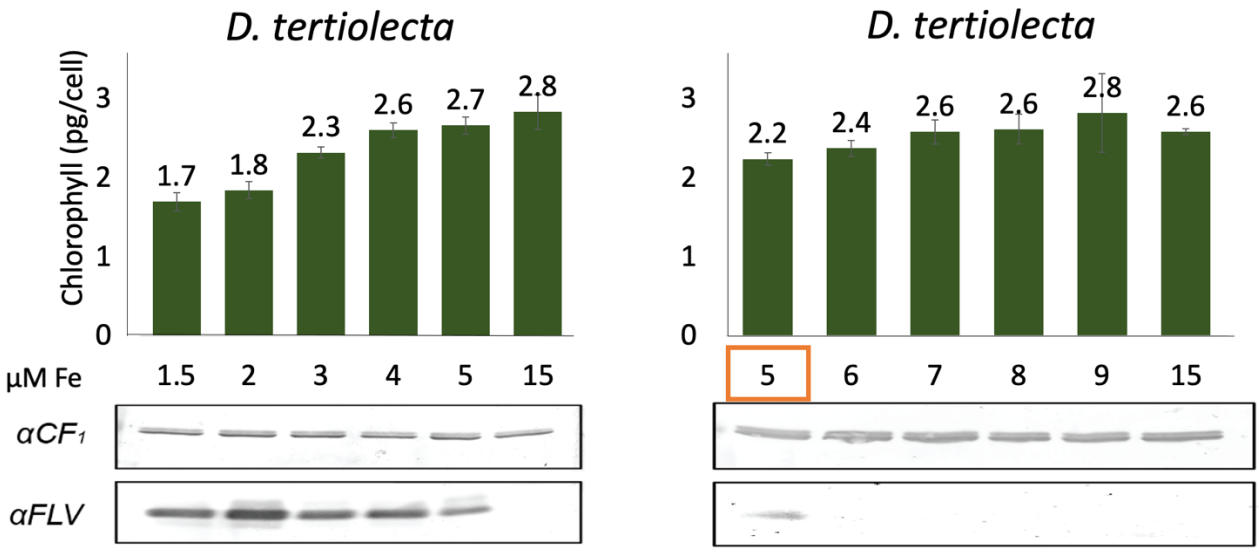

Figure 4. Chlorophyll content and immunoblots of FLV for *D. tertiolecta* under varying iron conditions. The iron deficient condition is boxed in orange.

To ensure reproducibility, I grew *D. salina* and *D. tertiolecta* once more in their determined iron replete, deficient, and limited conditions. I was able to verify that these conditions were correct and reproducible (Figure 5), with a significant decrease in chlorophyll content per cell in only the iron limited condition and the induction of flavodoxin in both the iron limited and deficient conditions. After I identified the iron limited, deficient, and replete conditions for *D. salina* and *D. tertiolecta*, I was ready to move onto analyzing how the photosynthetic efficiencies change across these iron conditions and across species.

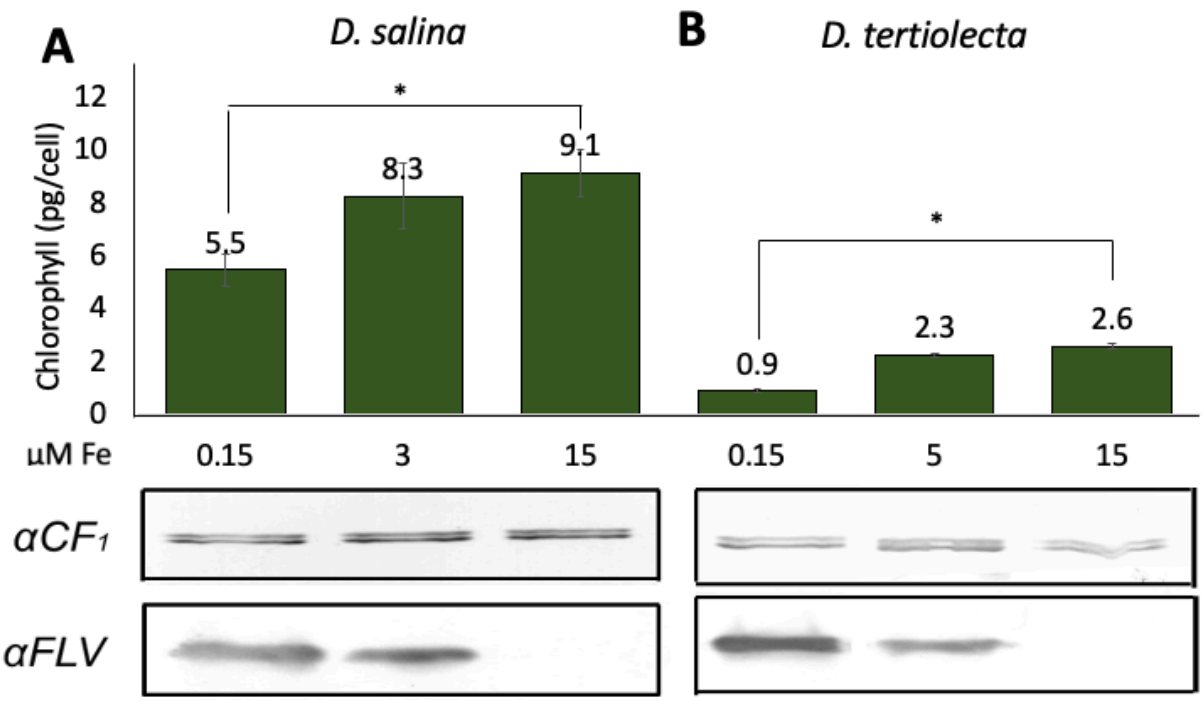

Figure 5. Chlorophyll content and immunoblots of FLV for (A) *D. salina* and (B) *D. tertiolecta* under their respective iron limited, deficient, and replete conditions. Significance was calculated using a t-test (p-value ≤ 0.05).

Comparing Photosynthetic Efficiency

To assess how the photosynthetic efficiency changes across iron conditions and across species, I grew new cells of *D. salina* and *D. tertiolecta* in their respective iron limited, deficient, and replete conditions and measured their F_v/F_m values, which is a measurement for Photosystem II efficiency. I was able to achieve an F_v/F_m value of around 0.7 for the iron replete conditions, which is the accepted efficiency value for Photosystem II efficiency. I found that there is a significant reduction of F_v/F_m in both *Dunaliella* species in the iron limited condition but not in the iron deficient condition when compared to the iron replete condition (Figure 6). Interestingly, *D. tertiolecta* has a much larger drop in F_v/F_m in its iron limited condition than *D. salina* does.

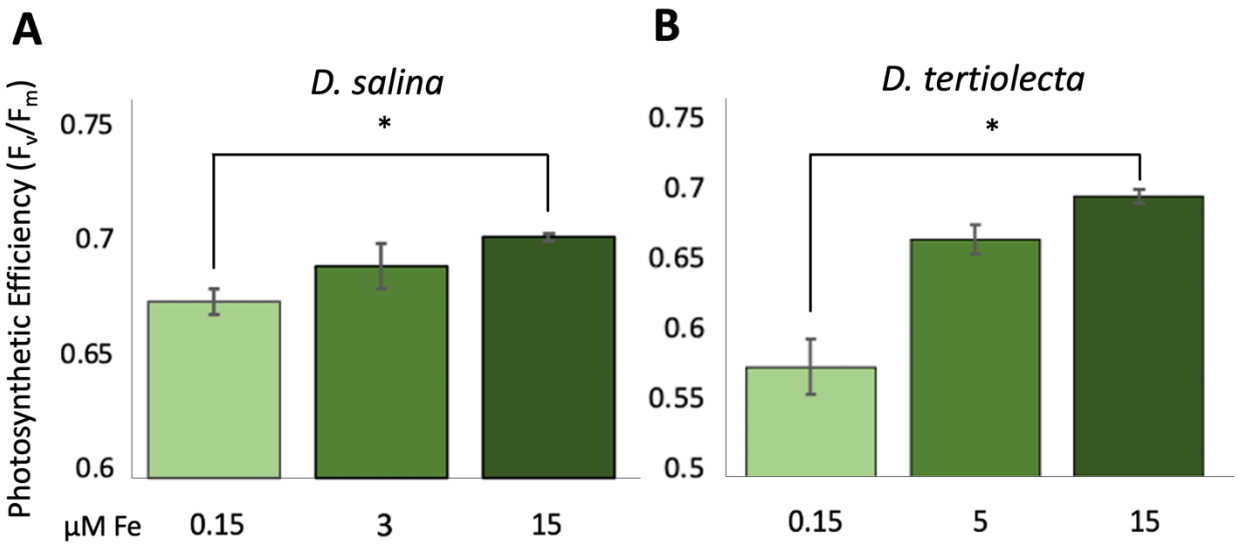

Figure 6. F_v/F_v values for *D. salina* and *D. tertiolecta* under iron limited, deficient, and replete conditions. Significance was calculated using a student t-test (p-value < 0.05).

DISCUSSION

From the results, I have determined the iron limited, deficient, and replete conditions for *D. salina* and *D. tertiolecta* and I have compared how the photosynthetic efficiencies of *D. salina, D. tertiolecta* differ across all three iron conditions. *D. salina* and *D. tertiolecta* have the same iron limited and replete conditions, however, the iron requirement for *D. salina*'s deficient condition is lower than that of *D. tertiolecta*. The F_v/F_m values for photoautotrophically grown *C. reinhardtii* are unaffected by iron nutrition, however iron limitation does decrease *D. salina* and *D. tertiolecta*'s F_v/F_m values, although to a lesser degree for *D. salina*. Further research needs to be done to make any definite claims. However, I have two main hypothesis which may explains these results.

Determining Iron Levels

I have determined the iron limited, deficient, and replete conditions for *D. salina* to be 0.15, 3, and 15 µM Fe and for D. tertiolecta to be 0.15, 5, and 15 µM Fe respectively. This result is surprising because literature values for *C. reinhardtii* has the deficient iron condition to be 2 µM Fe, which is lower than for both species of *Dunaliella*. Perhaps *Dunaliella* senses that it has low

iron availability at a higher concentration. Therefore, it can deploy its low iron mechanisms earlier to better survive in these low iron conditions. *D. salina* and *D. tertiolecta* are found in very salty environments while our strain of *C. reinhardtii* is a freshwater organism. Studies showed that when *Dunaliella* is grown in iron replete, high salt environments, it induces the iron uptake protein, p150. This suggests that high salt concentrations reduce the iron availability for *Dunaliella*, either through salt's effects on Fe^{3+} solubility or through an interference in its iron uptake machinery. Therefore, *Dunaliella's* higher iron deficient condition could be due to a combination of high salt stress and low iron stress.

However, *Dunaliella* also survives very well in media with no additional iron added. From the growth curves, we can see that even in the 0 μM Fe both species of *Dunaliella* are still able to grow. Typically, we expect to see reduced growth in iron limited conditions, which we do observe in *D. tertiolecta* (Figure 2). Interestingly, in *D. salina* we do not observe any effect of iron limitation on its growth rate.

Comparing Photosynthetic Efficiency

Under iron starvation, autotrophic *C. reinhardtii* grown in the absence of acetate maintains its F_v/F_m at 0.72 (Terauchi et al., 2010) while in *D. salina*, the F_v/F_m values drop from 0.70 to 0.69 to 0.67 in its iron replete, deficient, and limited conditions respectively and in *D. tertiolecta,* they drop from 0.70 to 0.67 to 0.58. The significant decrease in F_v/F_m indicates that the Photosystem II efficiency is decreased in iron limited *Dunaliella*. *D. salina'*s F_v/F_m values are less affected by iron limitation than *D. tertiolecta*'s values. One explanation of this could be due to *D. salina's* slower growth rate (Figure 7). Since *D. salina* grows slower and to a lower cell density than *D. tertiolecta*, it could be possible that there is more iron per cell in iron limited *D. salina* than in iron limited *D. tertiolecta.* This would mean that iron limited *D. salina* cells would be less iron stressed than iron limited *D. tertiolecta* cells and would therefore be able to maintain higher levels of photosynthetic efficiency. However, *D. salina* cells are much larger than *D. tertiolecta* cells. *D. salina* has a cell size diameter of around 14 microns while *D. tertiolecta* has a cell size diameter of around 8 microns. This size difference would increase the iron required by each *D. salina* cell and could therefore negate any affect growth differences might have on the level of iron stress *D. salina* and *D. tertiolecta* are experiencing under iron limitation.

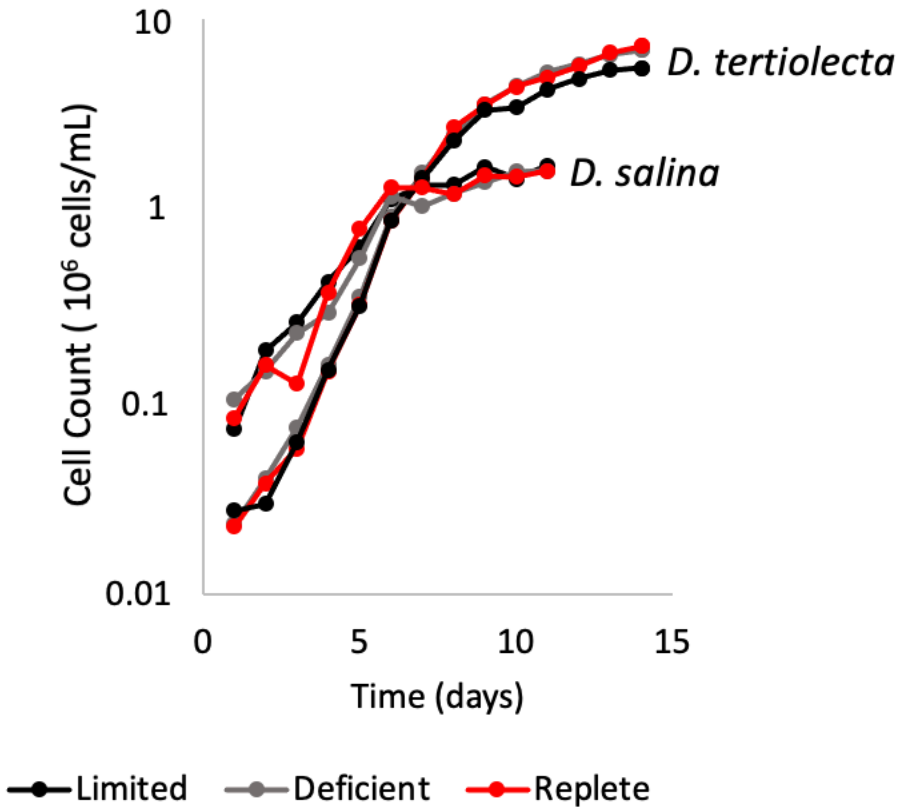

Figure 7. Growth rate comparison between *D. salina* and *D. tertiolecta* under their iron replete, deficient, and limited conditions.

Another potential explanation for why iron limited *D. salina* experiences a smaller reduction in F_v/F_m than iron limited *D. tertiolecta* is that *D. salina* could have a better low iron uptake system than *D. tertiolecta*. *D. salina* could induce high-affinity iron chelators, which could help it to bind to the small amounts of iron in the iron limited media. This would help *D. salina* to accumulate more iron per cell, which would help it to maintain higher levels of photosynthetic efficiency.

Limitations & Future Directions

There is still a lot of work to be done to further analyze the mechanisms which help *Dunaliella* to maintain its photosynthetic efficiency under low iron conditions. Some limitations of this study are that I was unable to analyze all the photosynthetic efficiency measurements I wanted, such as the analyze P700 oxidation and oxygen evolution and consumption rates, because project took longer than duration of the reporting period. This information would have been useful to analyze other components of photosynthesis, such as Photosystem I efficiency. In the future I

would like to analyze how these rates under the iron-limited, -deficient, and -replete conditions for *Dunaliella* change and how they compare to *C. reinhardtii.*

Additionally, another limitation is that I only analyzed *Dunaliella*, while there are many other species algae that are adapted to a variety of other environments. *Dunaliella* are halotolerant extremophiles, which means that they can survive in extremely high salt concentrations, up to salt lakes which are at full NaCl saturation (Oren 2014). Algae are found all around the world in other environments such as in freshwater, tree sap, and ice. These environments may affect the low iron mechanisms these other algae have, therefore studying numerous different algal species would provide valuable insights into their low iron mechanisms. However, it is still useful to study how *D. salina* and *D. tertiolecta*'s low iron mechanisms impact their photosynthetic efficiency under low iron conditions to be able to extrapolate how these other species algal species may function.

Understanding how the unique low iron mechanisms function in *Dunaliella* will give insight into engineering other photosynthetic organisms to respond better to low iron nutrition. This can increase agricultural crop yields because iron limitation in soils limits plant growth which affects arable lands worldwide (Zuo and Zhang 2010). This will not only help to combat world hunger, but also help to more sustainable grow crops and reduce the carbon footprint of the agricultural sector. Additionally, engineering more efficient plants and photosynthetic microorganisms will help with global primary productivity and oceanic carbon assimilation, which will help to remove CO_2 from the atmosphere. Algae have the potential to greatly aid us in the fight against climate change, so we need to promote algal research now more than ever before!

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