

Turn That Light Up: Examining the Effect of Light Intensity on Photosynthesis as Measured by Oxygen Production in *Elodea canadensis*

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Abstract:

Photosynthesis is an essential reaction occurring in all plants as it provides their source of energy. As light is one of the required reactants, we chose to investigate the effects of differing light intensities on the rate of photosynthesis, hypothesizing that greater light intensity will result in greater photosynthetic rate. The rate was measured by determining the change in oxygen concentration in the medium by the aquatic plant, *Elodea canadensis*. The plant was immersed in the medium and subjected to different light intensities over three independent trials. Although we were unable to find any statistical differences between change in oxygen concentration and the light intensity, a slight trend suggested light intensity did have an effect on the rate of photosynthesis. The effect of light intensity followed a Michealis-Menten curve where increases in light intensity corresponded to increases in oxygen production until a peak in oxygen production occurred at a light intensity of 6000 lux. From this, we suspect that *E. canadensis* has an optimal photosynthetic rate at light intensities of 6000 lux. At greater intensities oxygen inhibition begins to occur due to the ability of the enzyme ribulose phosphatase to act in both photosynthesis and cellular respiration. However, as our results were not statistically significant, further studies would have to be performed to determine the effect of light intensity on photosynthesis in *E. canadensis*.

Introduction

Photosynthesis is an important process in plants in which light energy is converted to chemical energy for life's processes that are necessary for survival of the plant (Karp 2008). The reaction occurs in the chloroplasts in the leaves of the plant (Nelson and Cox 2008). It is activated when a sufficient amount of light is absorbed by the chlorophyll pigments of the chloroplasts (Nelson and Cox 2008). These pigments will then activate subsequent enzymes which result in the photosynthetic reactions where carbon dioxide, water, and light are the reactants and glucose and oxygen are the products: $6\text{CO}_2 + 6\text{H}_2\text{O} + \text{light} \rightarrow \text{C}_6\text{H}_{12}\text{O}_6 + 6\text{O}_2$ (Nelson and Cox 2008).

We expect with increases in light intensity, more chlorophyll pigments will be activated, causing greater activation of subsequent enzymes in the photosynthetic pathway and therefore, resulting in greater production of glucose and oxygen. Rodrigues and Thomaz (2010), with a similar plant, *Egeria densa*, determined that increasing light intensity did increase the rate of photosynthesis. However, they found that this trend began to plateau at higher light intensities. Oxygen inhibition may explain this plateau (Ku *et al.* 1977) as the enzyme ribulose phosphatase which catalyzes photosynthesis, also catalyzes cellular respiration at higher concentrations of oxygen (Jordan and Ogren 1984).

We will examine the change in O₂ concentration as a measure of the rate of photosynthesis and determine the effects of light intensity on this rate in *Elodea canadensis*.

H_O: Increasing light intensity will decrease or will have no effect on the rate of photosynthesis in *E. canadensis*.

H_A: Increasing light intensity will increase the rate of photosynthesis in *E. canadensis*.

E. canadensis (informally known as “anacharis”) is a fast-growing freshwater aquatic plant native to North America and commonly known for its use in aquaria (GISD 2010). In the aquarium, the plant acts as décor but also contributes to increasing the oxygen concentration so the conditions are better for other plants and animals in the aquarium (GISD 2010). The plant grows as a long single stalk with bright green translucent leaves that circulate in groups of three (Figure 1) (GISD 2010).

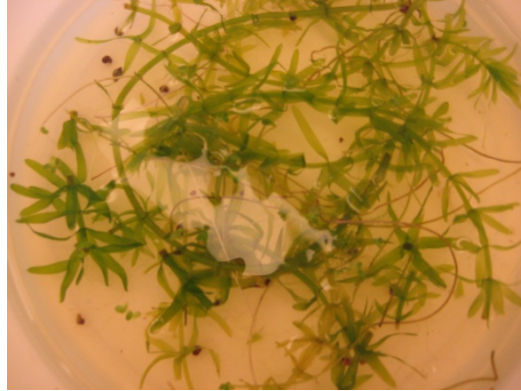


Figure 1. *E. canadensis* plant used in our experiment from Biology 140 aquarium.

We chose *E. canadensis* for its ability to photosynthesize while completely submerged under water so that all the products of its photosynthesis are released into the surrounding medium (GISD 2010). Therefore, any oxygen produced will remain in the medium (provided the plant is fully submerged) so we can more accurately measure oxygen concentration. In determining the effect of light intensity on photosynthesis in *E. canadensis*, we hope to determine the ideal conditions for the survival of this plant so it can be better utilized in aquaria.

Methods

We obtained live *E. canadensis* from an aquarium and weighed out the plant using a digital scale so we had fairly equal amounts in each treatment. We avoided cutting the plant as much as possible, aiming to have only one strand per treatment. We noted the initial appearance of the plant and the distribution of it when it was placed in the medium-filled jars. We used 110 mL jars filled with *Chlamydomonas* medium. When we were ready to begin the experiment, we added the *E. canadensis*. The jars had glass sides with a solid lid and each jar represented one replicate of one treatment. We had 6 treatments (each received a different light intensity), including a negative control (no light) and a procedural control (no organism added) with 3 replicates for each treatment. Treatment 1 was subjected to ambient light. Treatments 2, 3, and 4

were each under a 150 W flood lamp; all replicates of the same treatment would be under one flood lamp. We attempted to line the replicates in front of the lamp so that each replicate would receive a similar quantity of light. Since the lids of each jar were solid, we lay the flood lights on their side so the light would be directed at the glass sides of the jars. Treatment 2 received the full intensity of the lamp. The jars for Treatment 3 were wrapped once in cheesecloth to reduce the light intensity the plant would receive (Figure 2). Similarly, the jars for Treatment 4 were wrapped twice in cheesecloth to further reduce the light intensity the plant would receive (Figure 3).



Figure 2. Trial 2. Treatment 3. Jars wrapped once in cheesecloth.



Figure 3. Trial 2. Treatment 4. Jars wrapped twice in cheesecloth.

In Treatment 5, our negative control, we attempted to eliminate light as much as possible. The jars were wrapped twice in black plastic (Figure 4) and then all the replicates of this treatment were covered again by black plastic (Figure 5).



Figure 4. Trial 2. Treatment 5, negative control. Jars wrapped in black plastic.



Figure 5. Trial 2. Treatment 5. Black plastic was placed over all 3 jars to further reduce light.

Finally, Treatment 6, our procedural control was left at ambient light as in Treatment 1 but did not receive any *E. canadensis*.

In order to reduce the effect of changing light intensity with the time of day, we lined up the jars in a single row in an area where window light was minimal. The replicates of the same treatment were placed directly beside each other (in an order of Replicate 1, Replicate 2, Replicate 3) but the separate treatments were spaced out with 20 centimeters between each treatment. In total, we had 18 jars to be examined (Figure 6) with three replicates of each treatment.

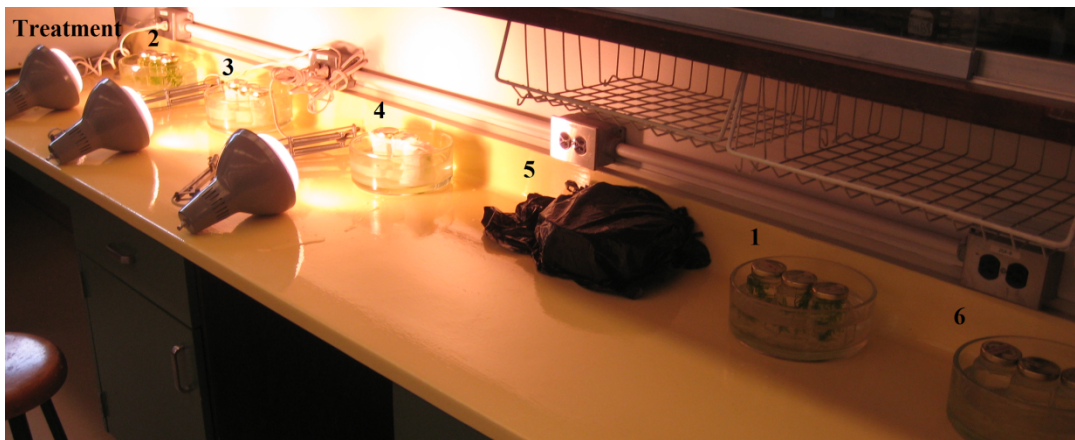


Figure 6. Trial 2. Treatments 1-6 set-up on a lab bench away from window light. Three replicates of each treatment.

For each treatment, we obtained oxygen concentration using a Vernier Dissolved Oxygen Probe. We took care to make sure that the probe did not interact with the plant or sides of the jar

and that it was inserted as far into the jar as possible without touching the bottom. Using a Fisher Traceable® Dual-Range Light Meter, we measured the light intensity of each treatment at the start and end of the experiment. For Treatment 1, 2, and 6, we placed the light sensor in front of the replicate (Figure 7) but for Treatment 5, we placed the light sensor under the garbage bag.



Figure 7. Trial 2. Measuring light intensity of the middle replicate using a Fisher Traceable® Dual-Range Light Meter of Treatment 2.

In Treatment 3 and 4, we draped the cheesecloth over the flood lamp (one layer for Treatment 3 and two layers for Treatment 4) and quickly placed the light sensor in front of the replicate before removing the cheesecloth. We also measured the temperature of each treatment. Throughout the experiment and at the end, we noted any changes in the plant's colour, shape, or size.

Trial 1

We used approximately 2.0 g (ranged from 1.99 g to 2.09 g) of *E. canadensis* in each jar with 90 mL of media. We obtained oxygen readings at the start of the experiment and every half hour for 2 hours for a total of five readings for each replicate in each treatment (90 readings total). Immediately after obtaining an oxygen reading, we obtained a temperature reading of the media in the jar. We did not make an attempt to control for temperature in this trial. We

measured the light intensity by placing the light meter sensor in front of the replicate in the middle of the light beam.

Trial 2

As in Trial 1, we used approximately 2.0 g (ranged from 1.92 g to 2.07 g) of *Elodea canadensis* in each treatment; however this time we used 110 mL of media to fill the jars and ensure that the plant was fully submerged. In this trial, we measured the oxygen readings only at the start and end of a 2 hour interval. We controlled for temperature in this trial by using water baths. Each treatment was placed in a water bath (6 water baths in total) with approximately 1200 mL of cold water. We then measured the temperature of the bath at the beginning of the experiment and checked it again every 10 minutes. We added ice as needed to keep the temperature of all the baths at around 23⁰C. We measured the light intensity of each replicate separately.

Trial 3

We used approximately 0.40 g (ranged from 0.33 g to 0.46 g) of *E. canadensis* in each treatment. As in Trial 2, we used 110 mL of medium in the jars; however, after the initial oxygen reading, we added approximately 10 mL more medium so that the jars would just overflow. To reduce oxygen loss, we placed saran wrap, with a small hole in it, over the lid of the jar after the initial reading. In our final reading, we removed the lid and inserted the oxygen probe into the small hole. We monitored temperature and measured light intensity as described in Trial 1.

Data Analysis

We determined change in oxygen by subtracting the initial value of oxygen concentration from the final value over the time interval for each replicate. In Trial 2 and 3, we did this over the 2 hour interval but in Trial 1, we did this over each half hour interval and then took the

average over all the half-hour intervals. Once we obtained the values for change in oxygen level, we subtracted the average oxygen change of Treatment 6 (procedural control) from all the treatments to calibrate for any changes of oxygen that may have occurred simply due to the environment and not the plant itself. Then, we calculated the change in oxygen per gram of plant material. Next, we averaged the replicates to obtain an average oxygen change for each treatment. To determine the average light intensity, we measured the initial and final light intensity of each treatment and averaged these two values. For Trial 2 and Trial 3, we measured the light intensity of each of the three replicates, averaging over three initial and three final readings. However, in Trial 1, we only measured once per treatment, averaging one initial and one final reading. Using Microsoft Excel 2010®, we graphed the average oxygen change for each treatment against the average light intensity of that treatment. We considered the change in oxygen to be the measure of the oxygen production by *E. canadensis*. Finally, we compared the 95% confidence intervals for the mean oxygen production for each treatment, assessing statistical significance; and looking for general trends.

Results

In Trial 1, we noticed the media did not sufficiently fill the jar even with the addition of the plant. In Trial 2, we added more medium so that it would fill the jar but some overflowed due to the insertion of the oxygen probe in our initial reading. Therefore, in Trial 3, we added even more medium after insertion of the probe so the jar was completely full for the course of the time interval. However, with the insertion of the probe for our final reading, media overflowed.

In Trials 1 and 2, we noted that the change in oxygen concentration was fairly high in some replicates at the lowest light intensity. We were concerned that the photosynthetic rate may have been maxing out so in Trial 3, we used a smaller amount of plant.

In Trial 1, we noticed a wide range of temperatures from 24⁰C in Treatment 5 to 38⁰C in Treatment 2. Once we controlled for temperature in Trial 2 and 3, the temperature remained fairly constant at about 23⁰C.

Throughout our experiment, in Trials 1, 2, and 3, we observed no changes in the morphology of the plant. The samples appeared to have the same colour, shape, and size at the initial measurement and final measurement. However, we noticed that there were variations among the plant samples in each jar: some had leaves spaced almost 0.5 cm apart with thinner leaves while some had broader leaves, clumped closer together. Moreover, some samples were darker green while some samples (usually the ones with thinner leaves) were lighter green. Additionally, despite all samples having a very similar mass, some had more leaves and less stem while others had more stem and less leaves. We observed that the stem weighed more than the leaves. In addition, the plants were not distributed evenly in the jars; some floated around while others stayed closer to the bottom. There were also small snails (< 0.5 cm in diameter) and clear fish eggs attached to some of the samples.

Sample calculation:

Using Trial 2, Replicate 1, 2, 3 (R1, R2, R3).

- 1) Oxygen change: Final concentration- Initial concentration

$$R1: (12.9 \text{ ppm}) - (7.7 \text{ ppm}) = 5.2 \text{ ppm}$$

$$R2: (9.2 \text{ ppm}) - (7.9 \text{ ppm}) = 1.3 \text{ ppm}$$

$$R3: (8.3 \text{ ppm}) - (7.3 \text{ ppm}) = 1.0 \text{ ppm}$$

- 2) Calibration with Procedural Control:

Subtract the average oxygen change of the Treatment 6= 1.67 ppm

$$R1: (5.2 \text{ ppm}) - (1.67 \text{ ppm}) = 3.53 \text{ ppm}$$

$$R2: (1.3 \text{ ppm}) - (1.67 \text{ ppm}) = -0.27 \text{ ppm}$$

$$R3: (1.0 \text{ ppm}) - (1.67 \text{ ppm}) = -0.67 \text{ ppm}$$

- 3) Calibration for Mass: Divide by Mass of each replicate

$$R1: 3.53 \text{ ppm} / 2.07 \text{ g} = 1.71 \text{ ppm/g}$$

R2: $-0.27 \text{ ppm} / 1.95 \text{ g} = -0.14 \text{ ppm/g}$
 R3: $-0.67 \text{ ppm} / 1.92 \text{ g} = -0.34 \text{ ppm/g}$

- 4) Average change in oxygen per mass for each treatment:
 $((1.71 \text{ ppm/g}) + (-0.14 \text{ ppm/g}) + (-0.34 \text{ ppm/g}))/3 = 0.41 \text{ ppm/g}$
- 5) Calculate the standard deviation:
 Using Microsoft Excel 2010® for Step 3
 $=\text{stdev}(1.71, -0.14, -0.34)$
 $= 1.13$
- 6) 95% confidence intervals (n=3):
 $1.96*(1.13/\sqrt{n}) = 1.28 \text{ ppm/g}$

Trial 1

Figure 8 illustrated the changes in oxygen concentration (oxygen production) at the average light intensity for each treatment.

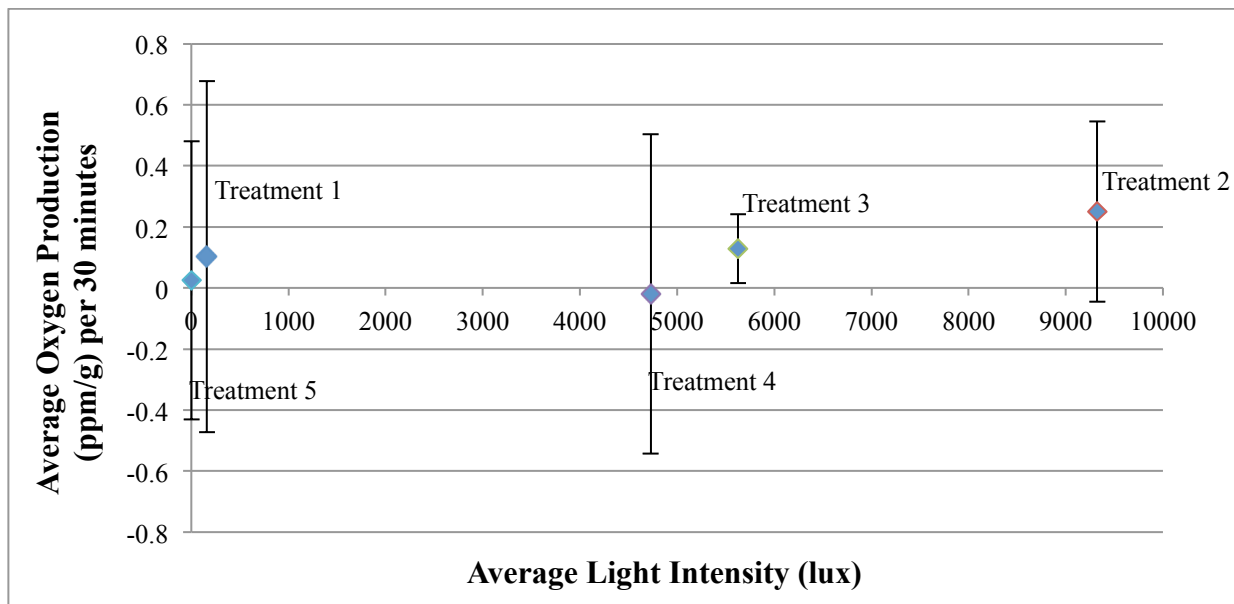


Figure 8. Effect of Light Intensity on Oxygen Production in *E. canadensis* (Trial 1). Oxygen production and is expressed per gram of *E. canadensis*. Change in oxygen was determined over five 30 minute intervals and averaged for each treatment, subtracting the value of a procedural control. Light intensity was averaged between the initial reading and the final reading. . Each treatment had 3 replicates. We analyzed the data with 95% confidence intervals.

The 95% confidence intervals of the mean changes in oxygen concentration (oxygen production) overlap (Figure 8), therefore the data suggests that varying light intensity does not have a statistically significant effect on oxygen production in *E. canadensis*. However, a slight trend (Figure 8) does appear to exist as change in oxygen increases with increasing light intensity in the following order: Treatment 5 (lowest light intensity, lowest oxygen production) < 1 < 3 < 2 (highest light intensity, highest oxygen production). A noticeable outlier in this trend is Treatment 4 which we would have expected to have a change in oxygen between Treatment 1 and 3 with this trend.

Trial 2

In total, we obtained five average changes in oxygen concentration (oxygen production) and the corresponding average light intensity for each treatment (Figure 9). See suggestion for figure 8.

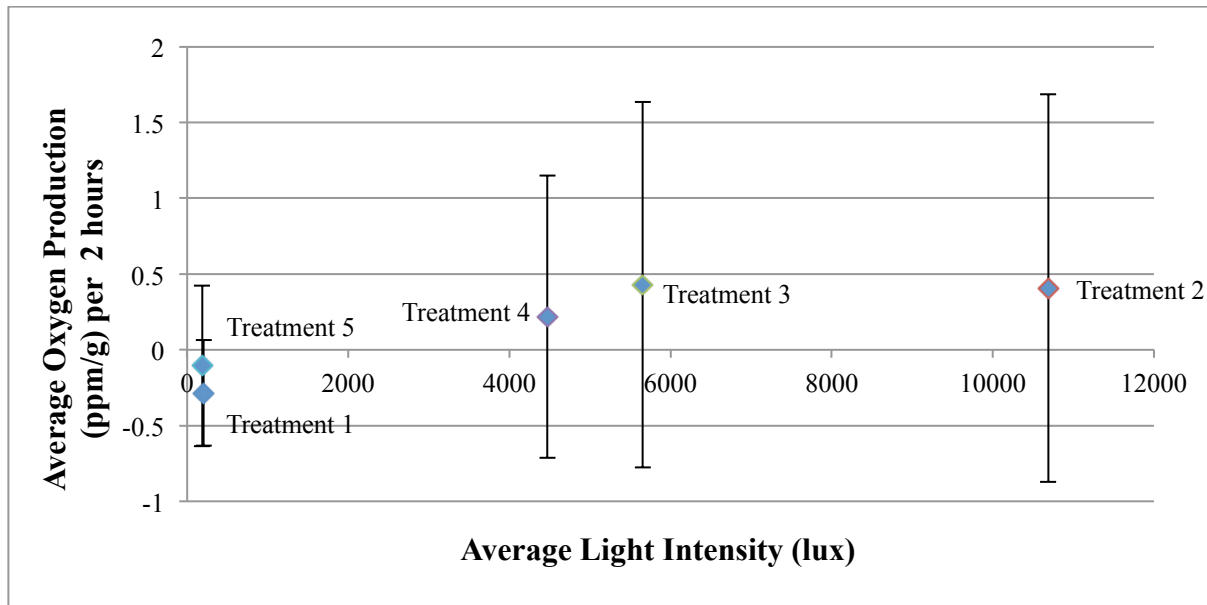


Figure 9. Effect of Light Intensity on Oxygen Production in *E. canadensis* (Trial 2). Oxygen production is expressed per gram of *E. canadensis* present in the medium. Change in oxygen of the procedural control was subtracted from each treatment. Light intensity was only averaged between the initial reading and the final reading after the final interval. Each treatment had 3 replicates. We analyzed the data with 95% confidence intervals. No statistical difference was found among average oxygen production in the treatments; however, a slight trend can be detected that increases in light do seem to lead to increases in oxygen production. This effect appears to plateau after a light intensity of 6000 lux.

Again, the confidence intervals of change in oxygen concentration overlap (Figure 9) among the treatments; therefore the data suggests that varying light intensity does not have a statistically significant effect on oxygen production in *E. canadensis*. However, a slight trend (Figure 9) does appear to exist as change in oxygen increases with increasing light intensity in the following order: Treatment 5 (lowest light intensity, lowest oxygen production) < 4 < 2 < 3 (highest light intensity, highest oxygen production). This relationship appears to be most dramatic before Treatment 3 at about 6000 lux; after this, the change in oxygen does not appear to increase much more with increasing light intensity. Treatment 1 is an outlier as we would have expected to have a change in oxygen between Treatment 4 and 5 with this trend.

Trial 3

The data from Trial 3 will not be considered as too little *E. canadensis* was used to provide detectable oxygen production. There was no detectable trend, nor statically significant relationships between oxygen production and light intensity; most of the changes in oxygen levels were close to zero for all treatments.

Discussion

We examined the effect of six different light intensities on the photosynthetic rate of *Elodea canadensis*. We measured changes in oxygen concentration and suggest that any increases in oxygen concentration is due to photosynthesis by *E. canadensis*. Our data show no statistically significant differences among the changes in oxygen at each light intensity treatment (Figure 8, Figure 9). Thus, we fail to reject our null hypothesis that increasing light intensity will decrease or have no effect on the rate of photosynthesis in *E. canadensis*. We fail to support our alternate hypothesis that increasing light intensity will increase the rate of photosynthesis in *E. canadensis*.

However, our data does show a slight trend. Although not statistically significant, it does appear that increasing light intensity results in increases in oxygen production (Figure 8, Figure 9). This trend appears largest at light intensities below 6000 lux; for light intensities higher than this value, the change in oxygen production remains fairly constant as if it had reached a maximum value (Figure 9).

This trend correlates well with the findings by Rodrigues and Thomaz (2010) who performed a similar experiment with *Egeria densa* (a plant related to *E. canadensis*) but on a much larger scale using approximately three times as much plant material and several trials with

a time interval from 34 to 37 days. They found that the relationship between photosynthetic rate in *E. densa* and light intensity follows a Michaelis-Menten curve (Figure 10) where it has a positive linear relationship initially but then the effect of light begins to decrease and the oxygen production becomes almost constant.

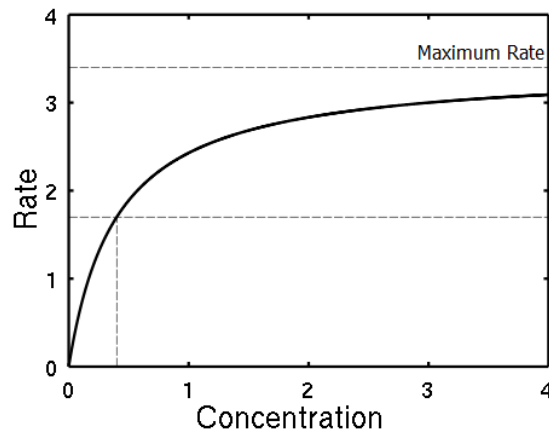


Figure 10. Michaelis-Menten saturation curve of an enzyme reaction. Image from Wikipedia.

Our graphs generally reflect this type of curve as the O_2 production largely increases below a light intensity of 6000 lux but above this light intensity, the O_2 production appears to become almost constant (Figure 9). It appears as if there is an optimal light intensity for *E. canadensis* where maximum photosynthesis occurs and after this light intensity, greater increases in light intensity will have very little effect. Under our experimental conditions, it seems as if this optimal light intensity occurred at 6000 lux (Figure 9).

As mentioned in the introduction, with increasing light intensity, the chlorophyll pigments will be activated at a greater rate, resulting in greater activation of the successive enzymes in the photosynthetic pathway (Nelson and Cox 2008). As a result, we expect that the concentration of oxygen will increase as more is produced by photosynthesis. This explains the earlier portion of our curve (Figure 9) as the change in oxygen appears to increase with light

intensity. However, it fails to provide an explanation for why the change in oxygen appears equal for light intensities beyond 6000 lux (Figure 9).

A potential reason why we see this trend was examined by Ku *et al.* (1977) in their examination of the plant *Solanum tuberosum*. They discovered that at a certain concentration of oxygen, the oxygen produced in photosynthesis by *S. tuberosum* actually began to inhibit photosynthesis itself. As a result, photosynthesis has a maximum rate before oxygen inhibition may begin to occur. In our study, this is particularly possible because we had our organism in a very small system (110 mL jar) and therefore, the produced oxygen cannot diffuse far and stays fairly concentrated. Oxygen inhibition occurring in *E. canadensis* at high light intensities may explain why the oxygen production becomes almost constant (Figure 9).

Oxygen inhibition likely occurs due to competitive inhibition of the enzyme ribulose phosphatase carboxylase (Bowes *et al.* 1971). The enzyme will catalyze the binding of carbon dioxide to ribulose-1,5-bisphosphate to form an intermediate that quickly breaks down into two molecules (glycerate-3-phosphate) (Nelson and Cox 2008). These products will be broken down in a series of steps, with the help of several different enzymes, to form the final products of photosynthesis (Nelson and Cox 2008) as described in the introduction. The rate of carboxylation by ribulose phosphatase is very slow and therefore, this step is the rate-limiting reaction of photosynthesis (Nelson and Cox 2008). However, ribulose phosphatase carboxylase can also bind to oxygen, acting as an oxygenase in the cellular respiration reaction (Nelson and Cox 2008). In cellular respiration, oxygen will be consumed in the following reaction: $C_6H_{12}O_6 + O_2 \rightarrow CO_2 + H_2O$ (Nelson and Cox 2008). Similar to its activity in photosynthesis, it will bind oxygen to ribulose-1,5-bisphosphate forming the intermediate products: phosphoglycolate and 3-phosphoglycerate (Nelson and Cox 2008). These products will also be broken down by the

action of several enzymes to form the final products of cellular respiration, producing energy for the plant's cellular activities in the process (Nelson and Cox 2008). Only one substrate can bind to ribulose phosphatase at a time so oxygen and carbon dioxide are competitive inhibitors of one another (Bowes *et al.* 1971). However, the enzyme will preferentially bind to carbon dioxide unless a high concentration of oxygen is present (Jordan and Ogren 1984). At first, little oxygen is present in the medium and therefore, ribulose phosphatase will primarily be binding to carbon dioxide and catalyzing the photosynthesis reaction. As the reaction proceeds, the carbon dioxide concentration begins to decrease and the oxygen concentration begins to increase due to photosynthesis. Eventually, with increases in oxygen, the enzyme will bind carbon dioxide and oxygen at the same rate so that the rate of photosynthesis equals the rate of cellular respiration. Therefore, the rate of oxygen production will equal the rate of oxygen consumption and the oxygen concentration will become nearly constant. This explains why above 6000 lux, there is very little change in oxygen concentration. In these treatments, so much oxygen was produced by photosynthesis that cellular respiration was occurring at the same rate and the oxygen concentration has plateaued. Eventually, as carbon dioxide is depleted by the carbon-fixing reactions of photosynthesis, ribulose phosphatase will bind oxygen at a greater rate than carbon dioxide and the rate of cellular respiration will actually begin to exceed photosynthesis. The oxygen concentration will begin to decrease and the change in oxygen may actually become negative. If we had left our experiment for a longer period of time, we may have seen this effect.

By looking at more light intensities closer to the optimal value, we would be able to determine where exactly this optimal value occurs and oxygen inhibition begins to occur.

Applications

As mentioned in the introduction, *E. canadensis* is a popular plant in aquaria, not only for its attractive appearance, but for its ability to photosynthesize while completely underwater. Therefore, it has the ability to supplement the aquarium medium with more oxygen which leads to better living conditions for the other plants and animals in the aquarium as oxygen is an essential factor for energy generation (Karp 2008). Our study attempted to determine the optimal light intensity for *E. canadensis* photosynthesis so its ability to produce oxygen can be enhanced. According to our overall trend, *E. canadensis* shows optimal photosynthesis (highest change in oxygen concentration) at light intensities of approximately 6000 lux (Figure 9). However, our results are only applicable to samples under our experimental conditions and we were unable to demonstrate that this value was statistically significant.

Biological Variation

Our samples of *E. canadensis* in each jar varied widely in their characteristics despite all coming from the same aquarium. We noted that some of the plant samples had slender leaves spaced almost 0.5 cm apart while some of the samples were much fuller with wide leaves closely clumped. From our data, we were unable to correlate which physiology resulted in higher or lower photosynthetic rates but Gifford and Evans (1981) found that the most successful crop plants, under direct sunlight, seemed to have broad, thin leaves. Therefore, the plants with the wider leaves may have a naturally higher photosynthetic rate, regardless of what light intensity they were exposed to.

Rascio *et al.* (1991) suggest another source of error is due to the structure of *E. canadensis*. They suggest that the leaf can utilize inorganic carbon in both forms as CO₂ or

HCO₃. while the stem, behaves as an aerial organ, requiring very high CO₂ concentrations in the medium in order to carry out photosynthesis. When we were selecting our samples some samples contained more stems while the others contained more leaves despite having the same mass. As a result, some of our samples may have had higher photosynthetic rates due to the presence of more leaves and less stem.

Moreover, one sample might have been older, or less healthy than others which could have reduced the photosynthetic rate. Our lack of oxygen production on Trial 3 may be due to this factor as the plant had already undergone several trials (in which it is removed from its natural environment for several hours) and perhaps, was not as healthy. The time for the plant to recover between trial 2 and 3 may not have been sufficient.

We noted that in some of our samples, we had small aquarium snails which would have consumed some of the oxygen produced by *Elodea canadensis*, reducing the change in oxygen. However, the effect was likely very minimal as there were very few snails and they were very small.

The possible biological variation can explain why we observed certain points (Figure 6- Treatment 4, Figure 7- Treatment 1) on our graphs which deviated from the Michaelis-Menten shape. These variations could be due to the fact that one sample may naturally have a greater photosynthetic rate than another sample and the difference is not just due to variations in light intensity. In our second and third trial, we did make attempts to ensure that the leaf morphology of all the samples was the same by visual inspection but it was very difficult to do so.

Sources of Error

Our plant samples were not all the exact same mass. The samples with more mass would have higher rates of photosynthesis due to the increased leaf surface to capture light. To correct

for this, we divided the oxygen produced by the mass of the plant. However, as noted by Gifford and Evans (1981), we should have measured the dry mass of the plant and divided the oxygen consumption by this value. This is because some plants may have contained more water due to their morphology. This would have caused their measured mass to increase so their calculated oxygen production would have been lower than it actually was. It would have been more accurate to measure the dry mass as done by Gifford and Evans (1987). This could explain why particular points on the graph were lower than expected and deviated from the Michaelis-Menten shape.

Oxygen loss was a huge concern in our experiment. To minimize it, we had chosen an aquatic plant that would release all of its oxygen into the medium, provided it was fully submerged. In Trial 1, we noted that the medium had not filled the jar completely, which may have resulted in oxygen becoming trapped in the air between the lid and medium. As a result, we would have lost any oxygen trapped there when we opened the jar for the final measurement, resulting in lower readings than expected. This could explain the low values in some of our data. In our subsequent trials, we added additional medium to prevent this from happening. However, we noted that with the insertion of the oxygen probe, we would lose medium as it spilled over the top of the jar. As oxygen would have been present in that medium, we likely lost oxygen with this spillage which would have resulted in measured values lower than expected. Additionally, the plant may have begun to consume the oxygen it produced in cellular respiration (Brown and Schwartz 2009). This means, the measured levels of oxygen may have been lower as the experiment progressed, resulting in lower averaged readings. We also noted that the distribution of the plant was not even throughout the jars. Depending on the placement of the oxygen meter,

we may have gotten higher readings due to interaction with the plant. We tried to avoid this but it was not always possible.

To measure light intensity in our experiment, we originally, in Trial 1, only took one measurement in front of replicate 2. However, we realized this would be inaccurate as each replicate likely received a different light intensity due to its positioning in front of the light. In our subsequent trials, we measured light intensity by placing the meter in front of each replicate and taking the average of these readings. Although this would be more accurate, we failed to measure around the different sides of the jars and it is very likely that each angle was receiving a different amount of light. Furthermore, we did not account for the effect of water and glass distortion of the light. We measured the light intensity out front of the jar but the glass of the jar would have refracted the light and the water; both in the medium itself and the water bath would have further refracted the light. Since we can't guarantee that all the jars were identical, the nature of this refraction may have caused different light intensities for each replicate so the light intensity on the actual plant may have been larger or smaller than the value measured.

Additionally, we averaged the light intensity not only over an entire treatment but over the course of the experiment between the initial and final readings. Although we did make attempts to distance the experiment from windows, the light intensity would still have varied with time of day and with electrical output of the lights. In future studies, it is advisable to measure more frequently to get a more accurate average light intensity.

We averaged changes in oxygen concentration among the replicates. We are assuming that the rate of photosynthesis remains constant over the course of the experiment (from initial to final reading) but this is likely not the case. As the experiment progressed, the concentration of

prerequisites for photosynthesis, CO₂ and water, would reduce as they are consumed. Therefore, with fewer reagents, the rate of photosynthesis may have slowed.

Ideally, our negative control should have received no light and therefore, would have a light intensity of close to 0 lux. We were unable to achieve this as we received light intensity readings varying from 2 lux to 4 lux. Our method of measuring the light intensity was not accurate as we measured it by placing the meter underneath the garbage bag that wrapped Treatment 5. Adjustments to the experimental method could have gained a more reflective reading (e.g. place the meter in a treatment 5 jar, seal lid, and wrap in garbage bag) that may have been closer to 0 lux.

After noting wide temperature variation in Trial 1, we controlled for temperature in Trial 2 and 3 but we did so by monitoring the temperature of the water baths used. We assumed that the temperature of the bath would be the same as the media within the jars but there was likely some difference.

Conclusion

We failed to reject our null hypothesis that increasing light intensity will decrease or have no effect on the rate of photosynthesis in *Elodea canadensis*. Therefore, we are unable to support our alternate hypothesis that increasing light intensity will increase the rate of photosynthesis in *E. canadensis*. However, we did observe a slight trend that increases in light intensity increase oxygen production up until an optimal light intensity of approximately 6000 lux but this trend was not statistically significant. After 6000 lux, the oxygen production remains fairly constant.

Acknowledgements

We thank Carol Pollock for her discussion and comments on our experimental design. We also thank Niki Holden, and Diana Rennison for answering our questions and making suggestions for our study. We are indebted to Mindy Chow for technical assistance and instrument facilities. We are also grateful to the University of British Columbia Biology Department for offering the Biology 342 course.

Literature Cited

- Bowes, G., Ogren, W. L., and Hageman, R. H. 1971. Phosphoglycolate production catalyzed by ribulose diphosphate carboxylase. *Biochemical and Biophysical Research Communications*, **45** (3): 716-722.
- Brown, M. H. and Schwartz, R. S. 2009. Connecting Photosynthesis and Cellular Respiration: Preservice Teachers' Conceptions. *Journal of Research in Science Teaching*, **46** (7): 791-812.
- Gifford, R. M. and Evans, L. T. 1981. Photosynthesis, Carbon Partitioning, and Yield. *Plant Physiology*, **32**: 485-509.
- Global Invasive Species Database (GISD). 2006. *Elodea canadensis* (aquatic plant) [online]. Available from <http://www.issg.org/database/species/ecology.asp?si=289&fr=1&sts=&lang=EN> [accessed 24 October 2011].
- Jordan, D. B. and Ogren, W. L. 1984. The CO₂/O₂ specificity of ribulose 1,5- bisphosphate carboxylase/oxygenase. *Planta*, **161**:308-313
- Karp, G. 2008. *Cell and Molecular Biology*, 5th edition. John Wiley & Sons Inc, Danvers, MA.
- Ku, S., Edwards, G. E., and Tanner, C. B. 1977. Effects of Light, Carbon Dioxide, and Temperature on Photosynthesis, Oxygen Inhibition of Photosynthesis, and Transpiration in *Solanum tuberosum*. *Plant Physiology*, **59**: 868-872.
- Nelson, D. L. and Cox, M. M. 2008. *Lehinger Principles of Biochemistry*, 5th edition. Palgrave Macmillan, New York, NY.
- Rascio, N., Mariani, P., Tommasini, E., Bodner, M., and Larcher, W. 1991. Photosynthetic Strategies in Leaves and Stems of *Elodea canadensis*. *Planta*, **185** (3): 297-303.
- Rodrigues, R. B. and Thomaz, S. M. 2010. Photosynthetic and growth responses of *Egeria densa* to photosynthetic active radiation. *Aquatic Botany*, **92** (4): 281-284.