# Emerging Investigators The effect of different concentrations of iron on the growth of Egeria (Elodea) densa

Alexander Hu, Stacey Holliday, and Steven Smith Pittsford Mendon High School, 472 Mendon Rd., Pittsford, NY 14534

### Summary

Iron-containing substances are known to affect the growth of many aquatic plants, such as Egeria densa. However, the concentration of these substances on the growth of E. densa was unknown to our best knowledge. We designed and performed an experiment to explore the effect of different iron concentration levels on E. densa. We observed that iron does play an important role in the overall growth of *E. densa*. We also found that there is a relationship between the amount of iron introduced and the growth achieved. The data indicates that (1) both plants in the iron solutions had more growth than the plant in the control tank (0.0% iron); (2) the plant submerged in the iron solution of 0.1% had a growth rate higher than the plant submersed in the 0.19% solution by a factor of two, approximately. We conclude that a ~0.1% iron solution is the optimal solution for the fast growth of E. densa.

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# Introduction

Egeria densa is a common aquarium plant that is often called the Brazillian Waterweed or the Larger-flowered Waterweed (1). In the aquarium trade it is sometimes referred to as *Anacharis*, or mistakenly as *Elodea densa* / canadensis (1). These, being separate species, have different preferences and growth characteristics. *Egeria densa* originates from South America and it prefers hard alkaline waters (1). It grows fairly fast and propagates quickly through fragmentation (1). Aquatic botanists have observed very fast growth in an aquarium setting (2). This species grows so well and propagates so easily that it is considered invasive in many areas (3). Other people have studied what effects the plant species *E. densa* has on its environment, such as the effect it has on nutrient density and phytoplankton population (4). It is important to study the growth factors of *E. densa* because of the significant impact it has on natural wildlife and ecosystems.

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Iron is important for transpiration in plants (iron is found in the enzymes controlling transpiration) and for the synthesis of chlorophyll. In *E. densa*'s natural habitat (North and South America) the iron levels can range from 0.5 milligrams per liter to 50 milligrams per liter, although the median is usually 0.7 milligrams per liter (5). It was believed that a higher content of iron would result in a high growth rate (1). However, to our best knowledge, no tests have been done on the effects iron has on growth. In general, ferrous substances are known to affect the growth of many aquatic plants. However, the concentration of these substances on the growth of *Egeria* was also unknown to our best knowledge. We

Day	Control	0.1% Fe	<u>0.19% Fe</u>
0	5	5.3	5
2	5.6	6	5.5
4	5.7	6.7	6.1
6	5.8	7.2	6.4
8	5.9	7.9	6.7
10	6	8.3	6.8
12	6.2	9.1	7.3
14	6.4	9.3	7.6
16	6.6	10.8	7.9
18	6.9	12	8.1
20	6.95	12.2	8.3
22	7	12.8	8.5
24	7.2	12.9	8.6

Table 1: Length of *E. densa* plants over time grown in the presence of iron. All measurements are in centimeters.



Figure 1: The experimental set up. (A) The plant in 0.19% iron solution, (B) The plant in the 0.1% iron solution, and (C)the plant in the control tank.

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Figure 2: Measured length of Egeria densa vs. time in three different solutions of iron chelate. Note the greater final length of the plants supplemented with iron. The 0.1% led to greatest growth. Orange lines have been superimposed on the graph to indicate when re-supplementation occurred. There are noticeable increases in length for the plant in 0.1% solution after each re-supplementation.

designed an experiment to understand the effect and the best concentration of iron for *Egeria densa*'s growth. The objectives of this investigation were to find a relationship between iron concentration and growth (if any) and to find the difference between the average growth rates (if any).

#### **Results**

Table 1shows the measured data from theexperiment. All lengths are in centimeters. The measuredlength of the plants in the experiment is plotted in Figure2against time (days) for the three cases: control (0%iron), 0.1% iron and 0.19% iron. The figure indicates that



Figure 3: A schematic graph helping to illustrate diffusion. Diffusion is when something, like water, moves from a high concentration to a low concentration. The dotted line represents a membrane in which substances can diffuse through.

the concentration of iron in the tank affects the growth rate of Egeria densa. The control had the lowest total growth and the lowest average growth rate. The 0.1% iron tank had the highest total growth and the highest average growth rate. The 0.19% iron tank had the second highest total growth and the second highest average growth rate.

Figure 3 is an illustration that portraits the diffusion process. This process may play a role in explaining the experimental results. Figure 4 shows the growth rate against time (days). The growth rate is defined as the height difference divided by the time duration. The growth rate spiked after re-supplementations. These spikes occurred roughly a day or two after re-supplementation.

**Figure 5** is a bar graph that displays the average growth rate for the three cases. The average growth rate and its standard deviation are calculated from the results shown in **Figure 4**. The standard deviation is displayed by an error bar for each case. The growth rates are 0.09±0.05 cm/day, 0.29±0.10 cm/day, and 0.14±0.05 cm/day for the control tank, the 0.1% iron tank, and the 0.19% iron tank, respectively.

### Discussion

The final overall length for the plants with iron supplementation was greater than the control. Thus, it can be concluded that iron had a positive relationship with overall growth length. However, it seems that the amount of iron in the solution was important to the growth length as well. The 0.19% iron solution yielded less growth than the 0.1% solution, although both still grew more than the control. This could be the result of diffusion. It is when something, like water, diffuses from a high concentration to a low concentration in an attempt to bring the solute

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**Figure 4:** The *Egeria densa* growth rate for each measurement period calculated from Figure 1. It is noted that growth rate increase occurs within two days of re-supplementation. Orange dashed lines have been superimposed on the graph to indicate when re-supplementation occurred. Growth rate is calculated by dividing the difference of length by the amount of Ln - Ln

time:  $\frac{L\pi - L_0}{T_{\pi} - T_0}$ . L<sub>o</sub> is the initial length at day T<sub>o</sub> and L<sub>n</sub> is the plant length measured at day T<sub>n</sub>.

concentration levels into equilibrium (see **Figure 3**). This diffusion might have pulled out too much water out of the Egeria densa cells, causing harm to them which in turn would slow down the growth. This seems the most plausible reason for the observed results. For example, the plant in 0.19% iron was observed as having brown and dying leaves, which may be an indicator of diffusion. In **Figure 4**, there are spikes in growth rate roughly corresponding to re-supplementation periods, when nutrients were put into the tanks. This suggests that the iron does have an effect on the growth rate of the plants supplied with nutrients.

**Figure 5** supports the claims that iron has an effect on growth. The average growth rates were calculated and standard deviation was plotted from this figure's data. The calculated average growth rates are 0.08 cm/day, 0.29 cm/day and 0.14 cm/day for the control (0.0% iron), the 0.1% and the 0.19% iron solution tanks respectively. On the graph the 0.19% iron solution has a lower growth rate than the 0.1% solution, although both still have a higher rate than the control (**Figure 5**). The growth rate in the 0.1% solution is greater than that in the 0.19% by over a factor of two.

The data collected from the experiment may, however, have some small errors. When the plants were picked up and measured on the same ruler, this may have mixed some of the water causing changes to the data. Segregation of supplies can eliminate this problem in the future. Disruption of developing root systems during measurement may have also caused some errors. Resupplementation may have also inadvertently increased the concentration of iron in certain tanks, thereby skewing the results. However, resupplementation was necessary because as the plants grew they took in iron, reducing the concentration, making it necessary to re-supplement the solution

The lighting quality may have caused limitations because the light may not have been equally distributed. Although all plants received light, the center tank, which was the 0.10% iron tank, may have gotten more light. This may have affected the growth observed.

To conclude, the concentration of iron is important to the growth of *Egeria densa*. Too much iron leads to a slightly decreased growth rate. It is indicated through the data from our experiment that a solution of 0.1% iron achieves the greatest growth rate. The 0.19% solution being the second over the control with no iron suggest that iron does indeed help, even if it has lower growth rate. Through this experiment we have learned the iron preferences of *Egeria densa*. Information obtained from this experiment can help to better control Egeria densa growth in both aquariums and the wild. It can also be used to further understand how to eliminate it from the wild, since it is an invasive species. Future experiments might consider using more plants in each tank to increase the accuracy in statistics.

#### Methods

Three one-gallon plastic tanks controlled at 21°C were used for the experiment. The tanks had gravel at

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**Figure 5. Egeria densa rate of growth in different solutions of iron chelate.** The standard deviation of each growth rate is overlaid on the graph. Average growth rate is calculated with information derieved from Figure 4. The average growth rate of the plant in the 0.1% iron tank had the fastest growth, two times greater than the rate of the o.19% iron plant. The control had the lowest growth rate.

the bottom to support the plants (Egeria densa). Each tank had one new shoot of E. densa (Figure 1). Each shoot was trimmed so they were around the same length at the start of the experiment. The plants were placed in the gravel. The light provided was an 18-inch, 15watt, 8,000 K All-Glass Aquarium fluorescent light bulb. The light was placed above the tanks. The plants were continuously exposed to the light for 12 hours each day. Each tank contained a different concentration of iron. The control tank contained no iron chelate. The other tanks had a 1 mL cup of solution in concentrations of either 0.19% iron chelate or 0.1% iron chelate poured into them. The solutions were marketed as "Flora Pride" and "API Leaf Zone", respectively. There was no difference between the two solutions besides the iron chelate concentrations. The other ingredient in both solutions was soluble potash (K<sub>2</sub>O), 3% for both. There was also water. The water and fertilizer solution in the tanks was re-supplemented with the same nutrient solution they received the first day for 2 weeks at the same time, every week, in order to replenish nutrients that may have been taken in by the plants. Measurements of each individual plant height were performed once every two days using a ruler. The length of plants was measured from base to the absolute tip to the nearest millimeter and recorded, out of the tank. The measured plants were then placed back into their respective tanks. The experiment had been conducted within a month. The data from the records were used to calculate growth rates.

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