

# Effect of Iron Presence on the Algae Growth

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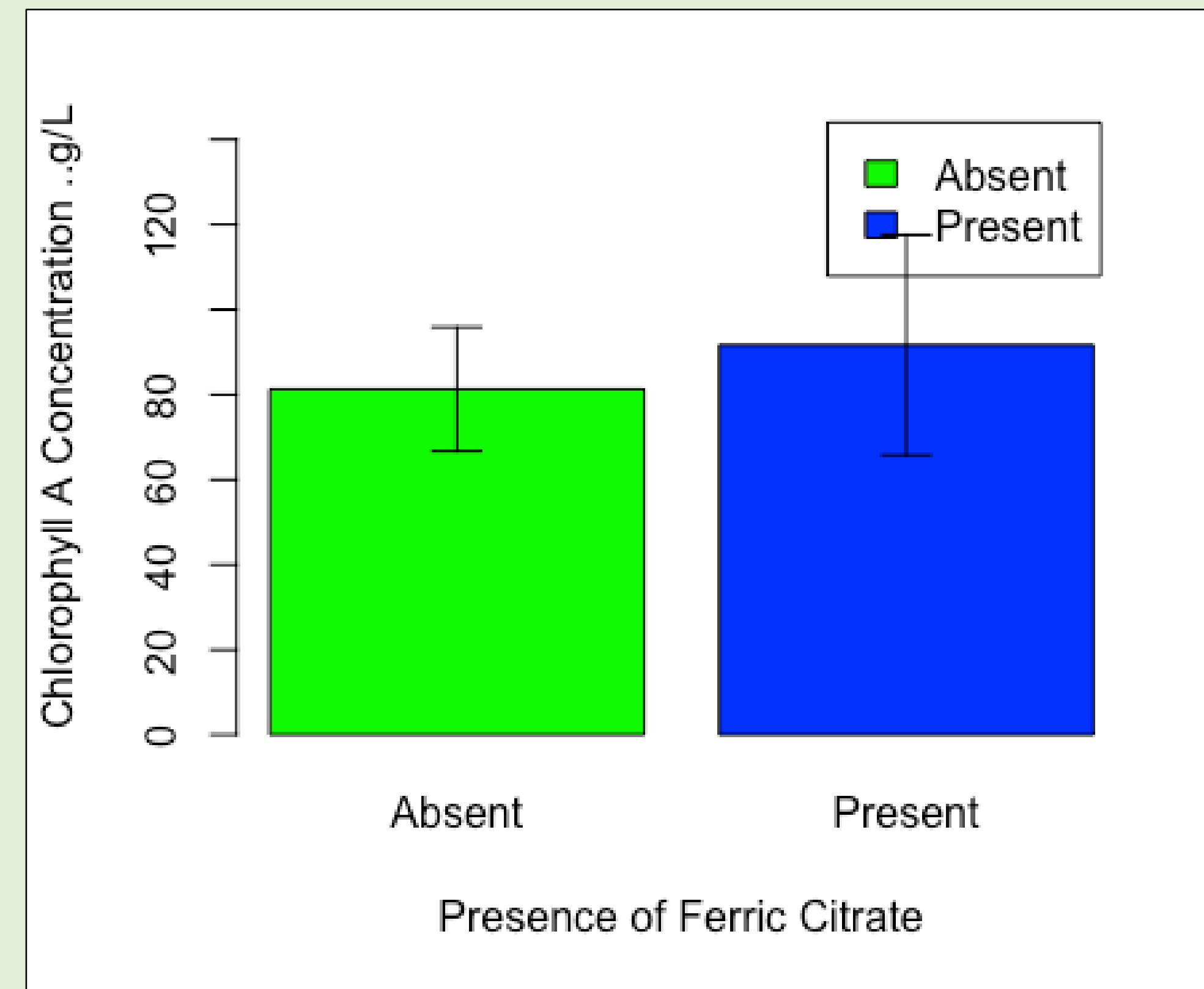
## Introduction

In this study, the effect of iron (in the form of ferric citrate) on the growth of algae was investigated. The uptake of iron in microalgae populations is of ecological importance and many microalgae species are able to reduce iron and store iron intracellularly. Iron is used by algae for metabolism and in the cellular processes of electron transfer and energy production (Sutak et al 2012). The iron requirements of different algae species varies, but generally, increased iron concentration is shown to increase algae density. Iron utilization is crucial for many aerobic organism's energy production processes which utilize the iron-rich photosynthetic electron transport chain (Sutak et al 2012). Overall, the medium of growth that algae takes place in, is of variable nutrient composition and has major impacts on algae population growth. In this study, it was hypothesized that the cell density of algae with the presence of Ferric citrate solution will be higher than that of algae without iron.

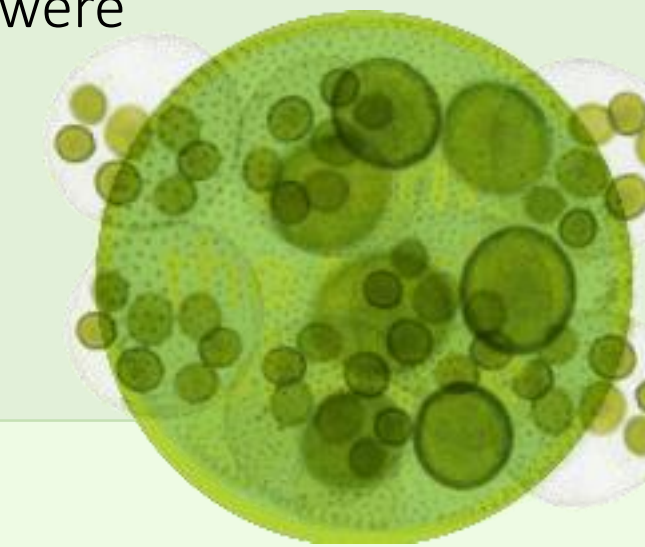
## Method

Ten pots, plastic cups and lids were obtained. The pots were sealed using masking tape and liquid agar to form a plug. Five clay pots were labelled "control" and five were labelled "treatment". 2% liquid agar was mixed with 1.00g ferric citrate and was added to the treatment pots. The control pots were filled to the rim only 2% liquid agar. The pots were placed in their labelled cups. 250mL of unfiltered lake water was added to each cup and closed with a lid. A week later, algae attached to the surfaces of the pot and cup was brushed off into the water in each cup. Eleven test tubes were labelled: five with "control", five with "treatment" and one with "blank". A cellulose filter was placed on the funnel, and the reservoir was placed to cover the funnel, and a vacuum was created with the faucet. For the blank, 100mL of deionized water was measured and poured into the reservoir. The reservoir and funnel were unclamped to obtain the filter membrane. The filter was cut into wedges and placed in test tube labelled "blank". This was repeated for all the samples; the sample solutions were used instead of water: 250mL of the control was used and 50mL of the treatment was used. Then, 10mL of 95% ethanol was added to each test tube. Each test tube was covered in aluminum foil and incubated at 75°C for 5 minutes.

The solutions were transferred into cuvettes and their absorbances were measured using Spectronic® 200. The concentrations of chlorophyll A was calculated from the absorbances the values, and were evaluated using Shapiro-Wilks Test and t-test.



**Figure 1.** Bar plot of mean average of concentration of Chlorophyll A, in micrograms per liter, against the presence of ferric nitrate. Data was obtained 7 days after initial contact with unfiltered pond water. Five trials were conducted for each mean value.



## Results

Since chlorophyll-a indicates the level of fertilization of a body of water, it can provide an estimate of algae concentration. Upon initial investigation, the control group had a lower mean chlorophyll-a concentration with a value of 81.27µg/L. Whereas, the treatment group had a higher mean chlorophyll-a concentration of 91.67µg/L. This suggests that the control group therefore experienced less algae growth in the same period of time of 7 days than the treatment group. Therefore, the micronutrient iron (used in the form of ferric citrate), most likely promoted the growth of algae. For the mean values, the Shapiro-Wilk test for normality was used to determine which statistical test to use. For both means, the p-value was greater than the chosen alpha value of 0.05. Therefore, they are normally distributed and the t-test can be used to compare the two samples of data. The p-value was 0.4174, which is greater than  $\alpha = 0.05$ , therefore the null hypothesis that there is no significant relationship between the presence of iron and algae growth is accepted and the alternative hypothesis is rejected.

## Acknowledgements

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## Discussion

The availability of limiting nutrient limits the productivity of algae (Elser, 2012). This study investigated whether the presence of iron (ferric citrate) acted as a limiting nutrient with the lake water to supplement the growth of algae. In the initial interpretation of the results, Figure 1 showed that the treatment group with the presence of ferric citrate appeared to have produced a slightly higher concentration of chlorophyll-a than that of the control group (absence of ferric citrate). However, upon statistical analysis, the correlation between the absence and presence of iron was not significant as the p-value is less than alpha level. ( $p = 0.4156 > 0.05$ ). Therefore, there was no statistical difference in algal growth between the treatment and control group. Although the hypothesis was supported that presence of ferric citrate has a higher cell density than that in the absence of ferric citrate, the presence of ferric citrate did not act as enough limiting nutrient to supplement the growth of algae. The lack of significance might be attributed to the limited supply of nutrient, and the restrained initial concentration of algae in the lake water. In order to improve the experiment in the future, the amount of targeted limiting nutrient should be increased, and initial sample of water should be better selected to have a higher concentration of algae. Also, possible experimentation to investigate iron as a limiting nutrients can also include varying the concentration of iron, so to determine the levels of iron required to produce a significant difference in algal growth.



**Figure 2.** Image showing the initial set up of the clay pots in the plastic cups.

## References

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- Sutak R, Botebol H, Blaiseau PL, Leger T, Bouget FY, Camadro JM, Lesuisse E. 2012. A Comparative Study of Iron Uptake Mechanisms in Marine Microalgae: Iron Binding at the Cell Surface is a Critical Step. *Plant Physiology*. 160(4):2271-2284.