

## **Analysis of Nitrogen Removal Efficiency of Advanced Integrated Wastewater Pond Systems (AIWPS)**

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**Abstract** Due to a lack of proper agricultural and municipal wastewater treatment facilities, a large number of the developing countries' population is still exposed to water pollution with limiting nutrients such as nitrogen. One of the promising technologies for wastewater treatment is Advanced Integrated Wastewater Pond Systems (AIWPS). The objective of this study is to determine nitrogen removal efficiency and its transformation pathways in AIWPS and to find out if water temperature, pH, and dissolved oxygen (DO) in AIWPS have the influence on those removal efficiency and transformation pathways. The two particular study systems of AIWPS are the Delhi system, which treats municipal wastewater, and the Panoche system, which deals with agricultural wastewater. Their capacity of nitrogen removal from those wastewaters was monitored over a period of 12 months. The Delhi system showed a relatively high total nitrogen and ammonia nitrogen removal of 78.9% and 75%, respectively over the whole period of investigation. On the other hand, the Panoche system removed only 44.6% of total nitrogen, and 56.5% of nitrate and nitrite. Highest removal of both of ammonia nitrogen, and nitrate and nitrite occurred during the warm period (28.3C° average), but the rates of removal of those forms of nitrogen were not correlated to pH and DO. The mechanism of relationship between water temperature and the removal of ammonia nitrogen, and nitrate and nitrite was discussed.

## **Introduction**

After carbon, hydrogen, and oxygen, the most abundant element in living cells is nitrogen. Thus the availability of various nitrogen compounds such as N<sub>2</sub> gas, nitrate, ammonium, and nitrite influences the variety, abundance, and nutritional value of animals and plants. In aquatic ecosystems the major forms of nitrogen available to aquatic creatures are nitrate and ammonia. They are, however, not always present in adequate amounts in natural waters and may limit plant growth, thus nitrogen is considered as one of the limiting nutrients. When there is excessive amount of nitrogen in aquatic systems, high nitrogen level in water gives enormous influences on aquatic environment (eutrophication) and on human health (blue baby) (Horn and Goldman, 1994).

Eutrophication can be defined as the process of enrichment of a water body due to an increase in nutrient loading (Horn and Goldman, 1994). A common indicator of this eutrophication is increased phytoplankton population density and speciation often with green, turbid, and foul smelling water and oxygen depletion in water. The source of nitrogen is the liberal use of fertilizers in agriculture combined with increased waste discharges especially after World War 2. Yet this fact that nitrogen is a source of pollution has not been well known by the public. Thus nitrogen sometimes is called the neglected pollutant (Horn and Goldman, 1994).

Human health can also be affected by nitrogen pollution. Throughout the world drinking water wells are contaminated (Cunte, 1997), exposing 3 million people only in the U.S. to nitrate contaminated water. The maximum concentration for nitrate of the human drinking water standard is 10mg/L, and of the people exposed above this level, approximately 43500 infants under about 6 months of age are at the risk of developing methemoglobinemia. This condition, which can lead to suffocation, occurs when nitrogen forms methemoglobin, which reduces the oxygen-carrying capacity of the blood (Horn and Goldman, 1994 and Magee, 1977). This oxygen starvation can produce a fatal condition known as blue baby syndrome. In the third world these problems have been becoming major concerns. Especially in China, the largest developing country in the world, still there are many people who lack access to safe waste supply and proper sanitation due to the lack of wastewater treatment facilities and intense use of land for agriculture, and fish farming (Lai 1994).

The treatment of nutrient-rich water has been very important for these reasons. Wastewater treatment systems such as High rate algal ponds are widely used for nitrogen removal around the world, and the efficacy of that system is well documented (Lai 1995). High rate algal pond

system is one of the most cost effective wastewater treatment systems, thus not only for the developing countries but also for other countries the application of high rate algal ponds seems to be a valuable way to treat nitrogen polluted waters for rehabilitation of the water body, sanitation and for reuse as irrigation and drinking water. However, there remains little understanding of the mechanics by which removal occurs or of the factors, which may affect the efficiency of the removal process (Cromar et al. 1996 and Reed 1985). Cromar and Reed stated that the efficiency of nitrogen removal is related to temperature and pH, especially in the summer. Hussanity (1979) also reported that total ammonium removal in particular nitrification, was higher during the summer. Contrary to those findings, Lai and Lam stated that nitrogen removal, nitrification, was more efficient during autumn and early winter, and dissolved oxygen (DO) level was strongly correlated with its removal. It seems there is still the need for further research on the relationship among nitrogen removal, pH, DO, and temperature in high rate algal pond system. The objective of this experiment is to find out the transformation pathways and removal rates of nitrate and nitrite and ammonia nitrogen in a wastewater treatment system, called AIWPS, Advanced Integrated Wastewater Pond System and to determine if the levels of pH, DO and temperature in the system have effects on nitrate and nitrite and ammonia nitrogen transformation pathways and removal efficiency.

## **Methods**

The purpose of this study is to measure how much total nitrogen is removed by AIWPS, and also it involves the determination of the effects of pH, DO, and temperature in AIWPS on removal efficiency of ammonia nitrogen and nitrate and nitrite. Since ammonia nitrogen is toxic to aquatic organisms and is the major component of nitrogen in one study site, Delhi system, ammonia nitrogen was used to determine correlation tests with pH, DO, and temperature. And also nitrate and nitrite were chosen to be used for the same correlation tests for the other study site, Panoche system because they are the source of nitrogen water pollution and the major component of nitrogen in this site. My hypothesis is that pH, DO, and temperature level have correlation with ammonia nitrogen and nitrate and nitrite removal efficiency and they act as independent valuable when those removal efficiency is dependent valuable. Under this assumption, pH, DO, and temperature levels are compared with mean monthly percentage nitrogen removal at each pond by linear regression test. This will show if there is correlation between those parameters and nitrogen removal rate.

**Data Collection** The study is carried out in a series of wastewater treatment ponds located at Delhi and Panoche, located on central San Joaquin valley, CA. Those ponds are called AIWPS developed by University of California, Berkeley's Engineering Field Station. AIWPS consists of a series of at least 4 ponds. In the Delhi system the first of the four ponds series is Advanced Facultative Pond (AFP) where municipal wastewater from the city of Delhi enters its digester pit where anoxic condition is kept. This anoxic zone is dominated by anoxic bacteria, which break down much of the sewage through methane fermentation and reduces biological oxygen demand (BOD). Nitrogen and phosphorous released by this activity are released to the oxic layer of AFP and taken up by algae. The second unit is high rate pond (HRP). This pond is designed to optimize algal growth by assimilation of CO<sub>2</sub>, NH<sub>4</sub>, and P available in water. At this stage most pathogenic bacteria are removed because of the high pH level caused by algal photosynthesis. In the next settling pond (SP) algae are settled out so that the treated water can be discharged to surrounding waterways. And the last one is maturation pond (MP), which once again disinfect the water and store it for irrigation. The Panoche AIWPS has a very similar structure. The first pond is reduction pond where oxygen is completely depleted and the perfect anoxic condition is made. Under anoxic condition nitrate is reduced to nitrogen gas, denitrification. The next pond is HRP. And lastly it enters SP or dissolved air flotation (DAF). They are algae harvesting ponds, and water is clarified.

Water samples are collected in a plastic bottle every Wednesday near the effluent pipe in each pond mentioned above, and the concentration of various forms of nitrogen and the levels of pH in the wastewater treatment ponds are measured in the Richmond Field Station (RFS) lab. DO and temperature are measured at the sites.

**Lab Analyses** *Organic Nitrogen Analysis:* Macro-Kjedahl Method was used to measure organic nitrogen concentration. There are three steps in this method. The first step is digestion. The sample was placed into an 800ml Kjeldahl flask and diluted up to 300ml with DI water. Using the tilting dispenser, 50ml of digestion reagent was poured into a digestion flask. Immediately after the pouring, the flask was swirled and heated, with the flask mouth over the blower. After all the water was boiled off, it begun to smoke. After a couple of minutes after the production of fumes, the flask was cooled for around 30 minutes. The second step is distillation. 50ml boric acid – mixed indicator solution (purple) was prepared in a receiving Erlenmeyer flask and placed under the distilling tubes. Using a tilting dispenser, 50ml of the sodium hydroxide-sodium thiosulfate solution (SHST) into the cooled Kjeldahl flask, which was diluted up to

300ml with DI water. Quickly the distilling tube stopper was placed into the flask mouth and swirled. It was heated until approximately 300ml distillate was collected into a receiving Erlenmeyer flask. The last step is titration. 0.02N HCl was added to the distillate flask. It was titrated until the original purple color of the boric acid indicator reappeared. Total Kjeldahl nitrogen, TKN, was calculated from the amount of HCl used for titration, and organic nitrogen was calculated from the equation as follows.  $TKN - NH_4 = \text{organic nitrogen}$ .

*Nitrite analysis:* Using the eppendorf repeater pipette (ERP), 10 L of color reagent was pipette into a sample, and it was sat for at least 10 minutes. The sample was mixed with the Dynatech holder, and the nitrate and nitrogen calculation machine called Dynatech calculated the concentration of nitrite.

*Nitrate analysis:* Using 250 L pipette, 250 L of the sample was transferred to the test tube, and 2.5mL of reagent was pipette into it with the ERP, and the test tube was mixed for 15 seconds with the vortex mixer. It sat for more than 10 minutes, and less than 1 hour to allow the color to develop, and was placed into a well. After this the concentration of nitrate was obtained by the Dynatech, which was used exactly the same way as the nitrite analysis).

*Ammonia analysis:* The 100mL sample in 150mL beaker was mixed, and the ammonia electrode and the temperature probe was immersed into the sample. 10N NaOH was added to the sample until pH was above 11. mV was measured with the electrometer.

*PH analysis:* measured at the site with a bench-top pH meter.

*DO analysis:* measured at the site using manufacturer probe membrane wrapped over electrode.

*Water Temperature:* measured at the site when DO was measured.

## **Results**

**Delhi system** Mean total nitrogen concentrations in each of four ponds at Delhi AIWPS, treating municipal wastewater, are shown in Figure 1. Raw sewage in the influent of the Delhi system contained about 53.9 mg/L of total nitrogen in an average of which, 78.1% (42.1 mg/L), was total ammonia nitrogen, while almost no nitrate and nitrite were present. Organic nitrogen was shown to have the largest treatment efficiency with >99% removal. High treatment efficiency of ON was observed particularly in AFP. The same holds true for ammonia, but for ammonia additional reduction in the HRP was observed. The ammonia nitrogen treatment

efficiency of Delhi system achieved 75%. Nitrate and nitrite production was observed in HRP (2.5mg/L).

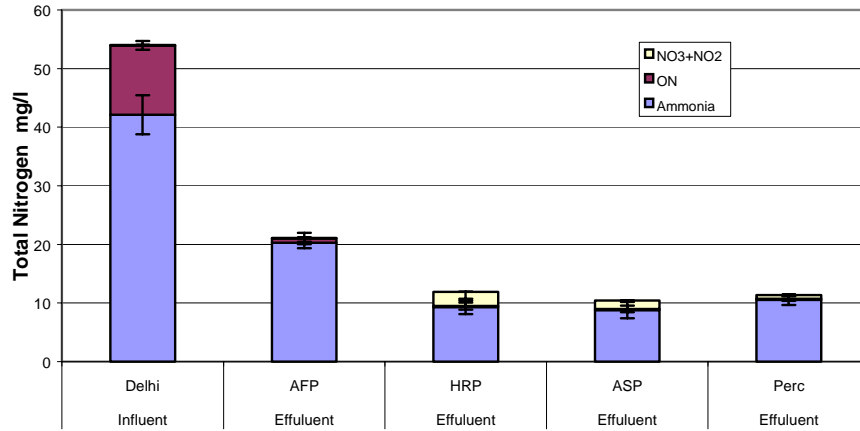


Figure 1: Concentrations of various forms of nitrogen in Delhi influent and those of effluent at the Delhi AIWPS. The figures show mean values (bar), standard error Total Nitrogen concentrations at the Delhi AIWPS over the period of investigation, from Feb 2000 to Feb 2001.

**Factors Influencing the Ammonia Nitrogen Removal** Linear regression tests were performed in order to test the effects of the average levels of water temperature, DO, and pH among the three ponds, AFP, HRP, and MP, on percentage ammonia nitrogen removal (Table 1). Statistically significant correlations between water temperature and ammonia removal efficiency in both of AFP and HRP were found, while DO and pH were not significantly correlated with the percentage removal of ammonia nitrogen. Based on these correlation results, ammonia nitrogen concentrations in warm period and cold period during the one-year investigation period in ponds in Delhi system were compared to see how actual water temperature affects the ammonia removal efficiency (Fig. 2). In warm periods at average water temperature 28.3°C ammonia nitrogen was removed through AFP and HRP, 63% and 88% removal efficiency, respectively, while relatively lower removal efficiency was observed in AFP

	AFP			HRP			MP		
Factors	R <sup>2</sup> -val ue	N	p-val ue	R <sup>2</sup> -val ue	N	p-val ue	R <sup>2</sup> -val ue	n	p-val ue
Temperature	R <sup>2</sup> = 0. 30	n = 20	<b>p = 0. 0127</b>	R <sup>2</sup> = 0. 54	n = 21	<b>p = 0. 0001</b>	R <sup>2</sup> = 0. 04	n = 33	p = 0. 283
DO	R <sup>2</sup> = 0. 16	n = 21	p = 0. 083	R <sup>2</sup> = 0. 001	n = 21	p = 0. 9921	R <sup>2</sup> = 0. 05	n = 31	p = 0. 244
PH	no pH data available								

Table 1: The correlations between the percentage removals of ammonia nitrogen in each pond in the Delhi system and possible factors that influence the removal efficiency during the one-year investigation Temperature, DO, and pH values for SP were not measured. pH for all the ponds was not measured. Statistical significances (p<0.05) are marked in bold font.

and HRP 42.4% and 41.8%, respectively in cool periods at average water temperature 11.6°C.

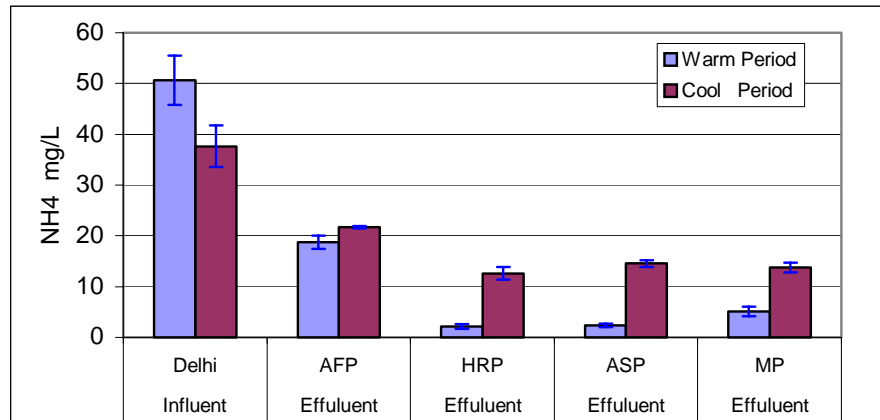


Figure 2. Comparison of ammonia nitrogen between warm period (Late May'00 to Sep'00) and cool period (Nov'00 to Feb'01) in the Panoche system. The figure shows mean values (bar) and standard errors for about the 16 samples for each pond.

**Panoche system** Mean total nitrogen concentrations in each of four ponds at the Panoche system, treating agricultural drainage water are shown in Figure 3. The influent of the Panoche system contained 65.4 mg/L of total nitrogen. Nitrate and nitrite were the dominating nitrogen fraction an average of 99.7% of the influent, 65.2 mg/L, whereas there was no ammonia in the influent and trace amount of ON, 0.2 mg/L. Through the Panoche system 56.5% of nitrate and nitrite were removed. A small amount of ammonia nitrogen was produced in the reduction pond, 0.57 mg/L, and reduced to virtually 0 mg/L in the effluent of settling pond. The production of organic nitrogen in HRP was observed.

**Factors Influencing the Nitrate and Nitrite Removal** Table 2 shows linear regression tests between percentage removal efficiency of nitrate and nitrite and water temperature, pH, and DO. A statistically significant correlation was obtained for the relationship between nitrate and nitrite removal efficiency and water temperature in HRP, while pH and DO were not significantly correlated with the percentage removal of nitrate and nitrite (Table 2). There is a steep decrease in nitrate and nitrite



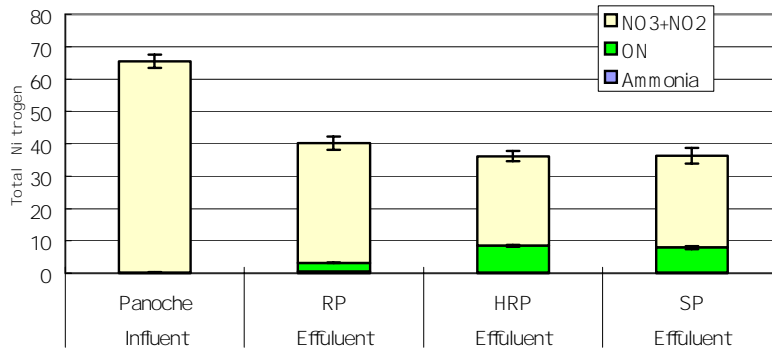


Figure 3: Concentrations of various forms of nitrogen in Panoche influent and those of effluent at the Panoche system. The figures show mean values (bar), standard error Total Nitrogen concentrations at the Panoche AIWPS over the period of investigation, from Feb 2000 to Feb 2001.

concentrations in HRP beginning late July until September 2000. The average temperatures and nitrate and nitrite concentrations for August and September are as follows, 27.5C: 14.13 mg/l and 26.6: 14.55mg/l, respectively. This interval, between July and September, was chosen as a warm period, and the same type of nitrogen removal analysis with water temperature as done for Delhi system was conducted for

Factors	RP		HRP		SP				
	R <sup>2</sup> -val ue	n	p-val ue	R <sup>2</sup> -val ue	n	p-val ue			
Temperature	R <sup>2</sup> = 0.02	n = 44	P = 0.413	R <sup>2</sup> = 0.40	n = 43	p < <b>0.0001</b>	R <sup>2</sup> = 0.002	n = 33	p = 0.851
DO	R <sup>2</sup> = 0.01	n = 46	P = 0.595	R <sup>2</sup> = 0.001	n = 43	p = 0.484	R <sup>2</sup> = 0.01	n = 31	p = 0.746
PH	R <sup>2</sup> = 0.01	n = 44	P = 0.440	R <sup>2</sup> = 0.01	n = 41	p = 0.448	R <sup>2</sup> = 0.03	n = 22	p = 0.585

Table 2: The correlations between the percentage removal of nitrate and nitrite in each pond in the Panoche system and possible factors that influence the removal efficiency during the one-year. Statistical significances (p<0.05) are marked in bold font.

the Panoche system (Fig. 4). In warm seasons 58% and 65.3% of nitrate and nitrite were removed through AFP and HRP, respectively, while relatively lower removal efficiency was noted in AFP and HRP in cool periods, 49.7% and 26.3%,

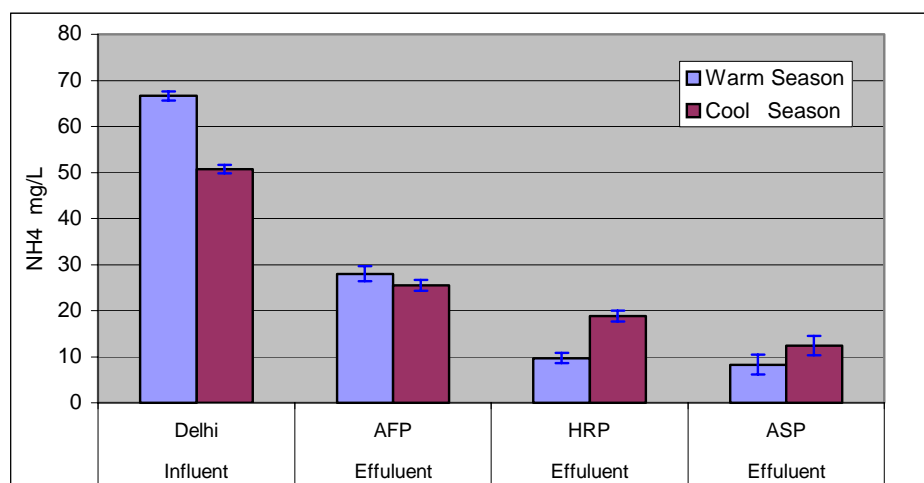


Figure 4. Comparison of nitrate and nitrite between warm period (Late May'00 to Sep'00) and cool period (Nov'00 to Feb'01) in the Delhi system. The figure shows mean values (bar) and standard errors for about the 16 samples for each pond.

respectively. There is an outstanding drop in removal efficiency, 39%, in cool periods in HRP.

## Discussion

In the Delhi AIWPS system showed relatively good removal of total nitrogen, 78.3%, while only 44.6% of total nitrogen was removed in the Panoche system. Delhi nitrogen removal values are commensurate with those found in the literature of between 54% and 96% (Cromer et al, 1996), while those of the Panoche system were below the above range. Ammonia nitrogen, the major component of nitrogen in municipal wastewater in the Delhi system, was removed, reaching 75% removal efficiency, and 56.5% of nitrate and nitrite in Panoche influent was removed through Panoche AIWPS system.

Temperature has been regarded as the most important physical factor influencing the efficiency of nitrogen removal because it directly affects the metabolic rate of micro-organisms and phytoplankton, algae (Gary, 1992 and Lai and Lam, 1995). Previous studies in Europe and America have shown that the nutrient removal efficiency by ponds was higher in summer than in winter (Pano and Middlebrooks, 1982; Santos and Oliveira, 1987). The present study followed the results of previous studies.

Removal of ammonia nitrogen and nitrate and nitrite was more efficient during warm period, which was from May to Sep'00. This tendency was shown mainly in HRP in both of the Delhi and Panoche systems. Ammonia is the preferentially utilized form of nitrogen for plant growth,

since the reductive metabolism of nitrate to the amino group  $\text{NH}_2$ , requires additional energy as well as the presence of an enzyme called nitrate reductase. This ammonia nitrogen was assimilated by the algae in HRP and contributed to the removal of ammonia nitrogen from wastewater. In the Delhi high rate pond, 51.9% of ammonia nitrogen from the previous AFP was removed. When there is no ammonia nitrogen, it is Panoche HRP in this case, algae take up nitrate as nutrients. Panoche HRP removed about 25% of nitrate coming in from the previous RP. There was statistically significant correlation between this ammonia nitrogen, and nitrate and nitrite removal efficiency and temperature in both of Delhi and Panoche HRP. Because of the seasonal high air temperature, water temperature in all of the ponds was also increased. This high water temperature stimulated algae in HRP, leading to high algal ammonia assimilation activity. One of my hypotheses that ammonia, nitrate, and nitrite removal is correlated with temperature was supported by the observed strong correlation in regression test.

However, no correlation between the nitrogen removal efficiency and dissolved oxygen and pH was found. During the period with possible high phytoplankton abundance (early summer to fall), enhanced photosynthetic activities of the phytoplankton not only increased the DO level, but also elevated the pH (< 9 pH average during this hot season) by consuming the acidic carbon dioxide in the high rate ponds. These conditions are optimal for the nitrifying bacteria and could have speeded up the rate of nitrification (Wild, 1971 and Lai and Lam, 1995). Lai and Lam also stated that nitrification was the principal removal mechanism of nitrogen. Actually in this study the production of nitrate and nitrite was observed in HRP in the Delhi system, possibly by nitrification, which occurred under the condition mentioned above, but whether this process contributed to ammonia nitrogen removal or not was not supported by the regression tests.

Since wastewater treatment systems such as AIWPS usually operate in relatively uncontrolled environment as compared to other mechanical treatment facility, the water temperature, pH, DO, solar insulation and phytoplankton population in the system changed with the weather (Maehlum and Stalnacke, 1999 and Lai and Lam, 1995). Thus the mechanism of this kind of wastewater treatment system is complicated and not easy to understand. If all or most of the factors mentioned in the previous page are related to each other, it is beyond the scope of my study. Also, loading rates and input concentrations of nutrients had an influence on the treatment efficiency (Maehlum and Stalnacke, 1999).

When all these factors are taken into consideration, my results may not be reliable. However, it is very possible to improve the quality of the study. First of all, the time that water samples,

pH, DO, and water temperature data taken should be fixed. pH, DO, and water temperature fluctuated throughout the day. Thus those data need to be collected at a certain time of a day. Some of my pH, DO, and temperature data were taken before sunrise, after sunset, and during the middle of the day, etc. And lastly, this kind of experiment should be repeated, or data from the past should be utilized to get more general ideas or results.

### **Acknowledgements**

Many thanks to Tryg Lundquist and Ingrid Xiviera at the Algae group at the Richmond Field Station in Richmond and Justin Victor Remais, Matt Orr, John Latto and Reuben Deumling for all your help.

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