



Published in final edited form as:

Water Environ Res. 2005 ; 77(7): 3003–3012.

Calcium Carbonate Phosphate Binding Ion Exchange Filtration and Accelerated Denitrification Improve Public Health Standards and Combat Eutrophication in Aquatic Ecosystems

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Abstract

Cultural eutrophication, the process by which a lake becomes rich in dissolved nutrients as a result of point and nonpoint pollutant sources, is a major cause of the loss of natural lake ecosystems throughout the world. The process occurs naturally in all lakes, but phosphate-rich nutrient runoff from sources such as storm drains and agricultural runoff is a major cause of excess phosphate-induced eutrophication. Especially in Madrona Marsh, one of the last remaining vernal marshes in the greater Los Angeles area, California, cultural eutrophication has become a major problem. In this study, calcium carbonate was found to be an excellent phosphate binder, reducing up to 70% of the phosphates in a given sample of water, and it posed relatively negligent ecological repercussions. This study involved the testing of this principle in both the laboratory and the real ecosystem. A calcium carbonate lacing procedure was first carried out to determine its efficacy in Madrona Marsh. Through this, ammonia was found to interfere with the solubility of calcium carbonate and therefore to be a hindrance to the reduction of phosphate. Therefore, various approaches for reduction of ammonia were tested, including aeration, use of fiber growth media, and plants, mainly *Caulerpa verticellata*, chosen for its hardiness, primarily in an attempt to increase population of *Nitrobacter* and *Nitrosomonas*. All were successful in moderately reducing ammonia levels. In addition, soil sampling, sediment analysis, microscopic plant analysis, microorganism and macroinvertebrate identification, and rate law formulations were conducted. The effect of phosphate and ammonia reduction on the populations of enterobacteria was also an important focus of this experiment. Varying concentrations of phosphate, ammonia, and calcium carbonate in conjunction with phosphate were tested in Madrona Marsh to determine their effects on the populations of enteropathogens on nonspecific blood agar, MacConkey agar, and Hektoen agar. Initial analyses suggest a strong correlation between phosphate concentrations and bacterial populations; a 66% decrease in phosphate resulted in a 35% reduction in bacterial populations and a 45% reduction in enteropathogenic populations. Likewise, a strong correlation was shown between calcium carbonate concentrations and bacterial reduction greater than that which can be attributed to the phosphate reduction alone. This was followed by the construction of various phosphate binding calcium carbonate filters, which used the ion exchange principle, including a spring loading filter, PVC pipe filter, and a galvanized filter. All were tested with the aid of Stoke's law formulation. The experiment was extremely successful in designing a working phosphate-binding and ammonia-reducing filter, and a large-scale agitator-clarifier filter system is currently being planned for construction in Madrona Marsh; this filter will reduce phosphate and ammonia levels substantially in the following years, bringing ecological, economical, and health-related improvements to the overall ecosystem and habitat.

Keywords

eutrophication; ion exchange; calcium carbonate; phosphate; phosphate binders; ammonia; denitrification; pathogens; enterobacteria; fresh water; lake; marsh

Introduction

Eutrophication is the process by which a body of water becomes rich in dissolved nutrients and minerals, markedly phosphate and nitrate compounds, and often grows shallow with a seasonal deficiency in dissolved oxygen, as a result of point and nonpoint pollutant sources. Eutrophication is a major cause of the loss of natural lake ecosystems throughout the world. There is also great potential threat to the survival of large lake organisms through oxygen depletion and toxin accumulation and risk for human health (Albering et al., 1999; Moody and Chu, 1995). This study involves the use of chemical and biological filtration and remediation to reduce the effects of eutrophication in two bodies of water in the greater Los Angeles area, California.

Harbor Lake Machado, one of the two bodies of water used for testing, is a moderately eutrophic lake in Los Angeles County and the largest natural lake in the greater Los Angeles area. It is a significant ecosystem, containing several unique species and attracting hundreds of migratory birds annually. The lake itself, though undergoing work by several environmental organizations, including the National Audubon Society and the Palos Verdes Peninsula Land Conservancy, continues to be polluted by various storm drains and nutrient-rich agricultural runoff. Surrounding areas, once home to various unique flora, are now vastly covered by non-native grasses. The storm drains and agricultural runoff from various locations drain water into the lake (Kao et al., 2002) and are most probably causing the recent rise in algae and bacterial growth (Kayombo et al., 2003).

This study also covered the nearby Madrona Marsh, a vernal marsh in the city of Torrance and the last vernal wetland ecosystem in the greater Los Angeles area that is of ecological importance. This is a very important migratory route along the Pacific flyway and houses a great diversity of organisms. Pollution is a major problem in Madrona Marsh. According to environmental impact statements from the 1970s (Del Amo, 1977), phosphate and nitrate levels are significantly above the standard safe limits (Keenan and Lowe, 2001). In addition, ammonia, chloride, bromide, iodide, copper, iron, and several other factors are predicted to be higher than environmentally acceptable limits because of the fact that Madrona Marsh is a continuing oil field, with six running oil wells in the center of the marsh. Within the marsh, there are many problematic sources of phosphates and other pollutants. A major pollution source in this particular preserve is the nearby carwash on Madrona Avenue near the northwest corner of the preserve. This water is extremely anoxic. Nearby households also generate a great deal of phosphate pollution (Del Amo, 1977). Much of this pollution reaches the marsh through the six storm drains and sump that feed into it.

Water from these two sources was used to test calcium carbonate (CaCO_3) as a potential phosphate binder (Legendre et al., 1994) to decrease the phosphate content in water. Phosphate binders, which are very common compounds in medicine used to control blood phosphate levels in many patients, especially those with hypertension (Ye et al., 2002), renal failure (Cannata Andia, 2000; Loghman-Adham, 1999), and renal insufficiency (Wen et al., 2003), include compounds such as aluminum hydroxide, calcium chloride, and calcium acetate (Lau et al., 1998; Nolan and Qunibi, 2003), among others, which are marketed as the pharmaceuticals Amphogel, Renagel, and Phoslo, respectively. Of these numerous compounds, calcium carbonate was chosen for many reasons, but especially because of its relatively large solubility

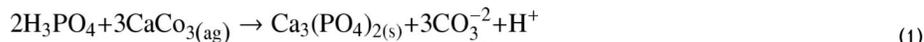
product (K_{sp}) in comparison with calcium phosphate [$Ca_3(PO_4)_2$] (Zumdaahl, 2000), but small in comparison to most other ionic compounds. In addition, calcium carbonate is relatively safe because of the natural decomposition of the carbonate ion. Calcium phosphate has a very low K_{sp} value of only 1.3×10^{-32} at $25^\circ C$, whereas that of calcium carbonate is 8.7×10^{-9} at $25^\circ C$. Calcium carbonate is also very inexpensive because it is a fairly ubiquitous compound; in fact, it is the largest reservoir for carbon. Filtration with calcium carbonate should be relatively easy compared to other compounds because calcium carbonate does not clump when exposed to water (Zumdaahl, 2000). Thus, these factors prompted the use of calcium carbonate in this study.

According to the U.S. Environmental Protection Agency standards and regulations (Billen et al., 1999), the safe levels of phosphates in any water system are not to exceed 0.1 ppm (3.2×10^{-7} mol/L). Waters containing higher phosphate concentrations are considered polluted. Dissolved oxygen standards hold that concentrations below 5.0 mg/L indicate anoxic conditions. Good water quality can be indicated by dissolved oxygen conditions of 7.0 to 9.0 mg/L. Nitrates higher than 10 ppm are extremely dangerous to the health of humans and other organisms found throughout freshwater lakes. Ammonium levels higher than 0.5 ppm are also indicative of poor water quality, and thus it is a goal to ensure the sustainability of sufficient nitrobacter and nitrosomonas populations to carry out the conversion of ammonia to nitrate (Keenan and Lowe, 2001). In this study, these measures have been used as gauges of water quality. In cases where the water quality parameters were not in agreement with the aforementioned standards, the water quality was deemed unacceptable.

This study examined calcium carbonate as a phosphate binder in aquatic situations, with an emphasis on the development of an ion-exchange filter with the conclusion of the study. Before the filters were constructed, various aspects of the calcium carbonate–aqueous phosphate reaction mechanism were considered, and the experimentation was modified accordingly. Auxiliary analyses, including soil sampling, sediment analysis, microscopic plant analysis, microorganism and macroinvertebrate identification, and parasite examination (Gelnar et al., 1997; Marcogliese and Cone, 1997) were also conducted to ensure that calcium carbonate would have insignificant detrimental affects on the aquatic ecosystem.

Hypotheses

Calcium carbonate is expected to adhere to the following reaction mechanism (eqs 1 and 2) and prove to be an effective phosphate binder, which acts to increase the dissolved oxygen levels, indirectly, in samples of water from fresh water lakes.



The typical photosynthetic pathway will improve the dissolved oxygen conditions on removal of phosphate and addition of carbon dioxide.

By adding small quantities of hydrochloric acid, which will decrease the pH, it is predicted that a greater quantity of calcium carbonate will dissolve in the water. Also, the second step of the reaction mechanism should become more rapid, thus also speeding up the final step. A maximal quantity of phosphate should also be removed with the lowest pH (Zumdaahl, 2000).

Temperature also affects the solubility of calcium carbonate. A lower temperature is best for its dissolution, as a maximal amount of carbon dioxide is able to dissolve in the water, thus speeding up the second step of the reaction mechanism and also lowering the pH. Ammonium chloride, an endothermically dissolving salt, was used in small quantities to lower the temperature (eq 3). There are, however, both negative and positive aspects of the use of ammonium salts to lower the temperature (Zumdaahl, 2000). While they are some of the safest endothermic salts, they are still harmful to living organisms.



Ammonia, because of its basic nature, is hypothesized to inhibit the dissolution of calcium carbonate (Zumdaahl, 2000). Because of its harmful nature to living organisms in excess, it is also beneficial to remove it from the water. Therefore, various ammonia reduction methods, including aeration (Burgess, 2003) and bacterial growth fiber medium were tested.

Also in this experiment, various filters, including a spring-loading design, a PVC design, and a galvanized steel design, was fitted with varying forms of calcium carbonate to test for efficacy in removing phosphate from a given sample. The spring-loading filter, because of its larger surface area and smaller diameter, in accord with Stoke's Law (Yamada, 2000), will probably produce the filtrate with the lowest phosphate concentrations, while the PVC design will probably produce the filtrate with the highest phosphate concentrations. However, the water will probably take the longest to travel through the spring-loading filter. The basic model for these filters is shown in Figure 1.

Methods

Lake Machado Analysis

An initial analysis was first started to determine relative pollutant levels at Lake Machado. Five identical containers were filled with one liter of water from the control sample or a storm drain at Lake Machado. The first container, used as a control, was filled with lake water from a source not within close range of the storm drains that feed into the lake. The second was filled with a simulated agricultural runoff containing one liter of lake water with two grams of dissolved Plant Gro fertilizer (Flora Care Products, PetCo, San Diego, California), simulating the water that actually runs into the lake from surrounding fields. This measurement was determined by analyzing the average amount of fertilizer placed on the lawn and the average amount of water used in irrigating the lawn. The other three containers were filled with one liter of lake water from the three storm drains that feed into the lake. All containers were placed in an area of light to moderate indirect sunlight to simulate the natural conditions. All water quality testing was conducted using the LaMotte SMART Colorimeter (LaMotte Company, Chester-town, Maryland). The water was tested every two days for phosphate using the LaMotte Ascorbic Acid Octet Comparator/ Axial Reader (3121/PAL) and dissolved oxygen content with a LaMotte Direct Reading Titrator (7414/EDO). The salinity of the water and specific gravity of the water, used for determining the relative evaporation, were tested using a LaMotte hydrometer (30025). Temperature was tested for relative fluctuations using the LaMotte non-mercury Armored Thermometer (range -25 to 45°C) (1066). Testing of water samples continued for 20 days.

Initial Calcium Carbonate Testings

A second analysis followed to test the efficacy of calcium carbonate as a phosphate binder in water collected from Lake Machado. Five identical containers were filled with one liter of water from a specific source in the lake. The first container, used as a control, will have no

chemicals added to it. The second container had 15 grams of calcium carbonate (Sigma-Aldrich Company, St. Louis, Missouri) added as a phosphate-binding compound. The third container contained calcium carbonate with ammonium chloride (NH_4Cl) (Sigma-Aldrich Company), used as an endothermically dissociating agent. The fourth container contained the calcium carbonate along with the aquatic plant, *Caulerpa verticellata*. The final container had all three—calcium carbonate, ammonium chloride, and the plant—added to the water. Then, one milliliter of 1-M hydrochloric acid (Sigma-Aldrich Company) was added to containers two through five. Ammonium chloride was used in the small quantity of 5 grams per 1.5 liters, a concentration of only 0.06 mol/L. All containers were placed in an area of light to moderate indirect sunlight to simulate the natural conditions. The water was tested every two days for phosphate using the LaMotte Ascorbic Acid Octet Comparator/Axial Reader (3121/PAL) and dissolved oxygen content with a LaMotte Direct Reading Titrator (7414/EDO). The salinity of the water and specific gravity of the water, used for determining the relative evaporation, were tested using a LaMotte hydrometer (30025). Temperature was tested for relative fluctuations using the LaMotte non-mercury Armored Thermometer (range -5 to 45°C) (1066). Testing of water samples continued for 20 days.

Madrona Marsh Analysis

Water samples from Madrona Marsh were then collected to test for various water quality parameters, including phosphate, nitrate, dissolved oxygen, ammonia, chromium, iron, copper, and chlorine/chloride. All parameters were measured using the LaMotte SMART colorimeter. Phosphates were tested using the ascorbic acid reduction method (3653-SC); nitrates were tested using the nitrate-nitrogen cadmium reduction method (3649-SC); dissolved oxygen was tested using the Winkler colorimetric method (3688-SC); ammonia was tested using the high range ammonia-nitrogen nesslerization method (3642-SC); chromium was tested using the Diphenylcarbohydrazide method (3698-SC); iron was tested using the Bipyridyl method (3648-SC); copper was tested using the high range Diethyldithiocarbamate method (3646-SC); and chlorine was tested using the DPD method (3643-SC).

Calcium Carbonate Reaction Rates

In addition, laboratory rate law formulation of calcium carbonate reacting with aqueous phosphate was conducted using pH balanced sodium phosphate solutions (Sigma-Aldrich Company). Differing phosphate concentrations were treated with differing amounts of a saturated calcium carbonate solution to find a minimum amount of calcium carbonate needed to reduce a maximal amount of phosphate. The same study was repeated with differing pH-balanced concentrations of ammonium chloride and sodium phosphate to assess the effects of ammonia concentrations on calcium carbonate. In a third stage, aqueous basic ammonia of 5% concentration (Sigma-Aldrich Company) was added in specific aliquots to a pH-balanced sodium phosphate solution to test the effects of pH varying ammonia on calcium carbonate solubility and its efficacy in reducing phosphate concentration in the water.

Calcium Carbonate Lacing Procedure for Phosphate

Three 1-m^3 volumes of water were sequestered by barriers in the sump, which supplies Madrona Marsh. This measurement was approximated using exact length and width measurements and approximated depth measurements with each area having a one-meter edge length at the edge of the sump. The first area was not treated and left to remain “as-is” throughout the entire duration of this phase. The second area was treated with 20 g of calcium carbonate dispersed throughout the delineated region. The third region was treated with 20 g of calcium carbonate and 10 mL of concentrated hydrochloric (muriatic) acid (33.5% HCl). The hydrochloric acid, in small volumes, does not harm the organisms in the marsh, but aids in the dissolution of the calcium carbonate. The HCl and the CaCO_3 neutralize each other, and,

therefore, no noticeable pH change should be observed. Measurement of all of the aforementioned parameters (phosphate, nitrate, dissolved oxygen, ammonia, chromium, iron, copper, and chlorides) was taken immediately before and after treatment. Phosphate, pH, and dissolved oxygen were measured weekly, and the other parameters were measured biweekly. After four weeks, the barriers were removed and the water allowed to equilibrate.

Sediment Analysis

The sediments from all analyses and each of the delineated areas in the lacing procedure were filtered using standard-grade filter paper (VWR International, Inc., West Chester, Pennsylvania). The additional water was boiled using a hot plate (VWR International, Inc.) and a Bunsen burner (VWR International, Inc.), until the level reached 25 mL, according to common sediment analysis protocol (Cannata Andia, 2000). This water was then tested for the presence of phosphate and calcium ions. The sediments were tested for calcium phosphate and calcium carbonate. This gave a clear image of the purity of the water and sediment after being treated with calcium carbonate with the addition hydrochloric acid and/or ammonium chloride.

Ammonia Approach Control Phase

Five identical containers were filled with 1500 mL of distilled water. To each container, a 5-ppm aliquot of ammonium chloride was added. In addition, the concentration of phosphate was varied in each container, from 0 to 10 ppm at a 3:2 mixture of sodium phosphate and potassium phosphate (Sigma-Aldrich Company). Nitrate was consistently added at 15 ppm in the form of sodium nitrate (Sigma-Aldrich Company) to each container to match the standard nitrate concentrations in Madrona Marsh. The first container was designated as a control. The second container was aerated daily for 10 minutes. The third was fitted at its base with a fiber media (PetCo) to increase the surface area for growth. The fourth and fifth containers were replicates of the second and third containers, respectively, with the exception of the added *Caulerpa verticellata*. All containers were placed in an area of light to moderate indirect sunlight for one week to simulate the natural conditions. The water was tested periodically for phosphate, ammonia, nitrate, pH, chlorides, and dissolved oxygen content. All parameters were measured using the LaMotte SMART Colorimeter. The salinity of the water and specific gravity of the water, used for determining the relative evaporation, were tested using a LaMotte hydrometer (3-0025). Temperature was tested for relative fluctuations using the LaMotte nonmercury Armored Thermometer (range -5 to 45°C) (1066).

Ammonia Approach Marsh Analysis Phase

Five identical containers were filled with 1500 mL of water from Madrona Marsh. The first container was designated as a control. The second container was aerated daily. The third was fitted at its base with a fiber media. The fourth and fifth containers were replicates of the second and third containers, respectively, with the exception of the added *Caulerpa verticellata*. All containers were placed in an area of light to moderate indirect sunlight for one week to simulate the natural conditions. The water was tested periodically for phosphate, ammonia, nitrate, pH, chlorides, and dissolved oxygen content. All parameters were measured using the LaMotte SMART Colorimeter. The salinity of the water and specific gravity of the water, used for determining the relative evaporation, were tested using a LaMotte hydrometer (3-0025). Temperature was tested for relative fluctuations using the LaMotte nonmercury Armored Thermometer (range -5 to 45°C) (1066).

Microscopic Analysis of Plant Specimens

The plants, which were used in the experiment during the initial analyses and the ammonia approach stages, were examined, first under a dissecting microscope (VWR International, Inc.), and then under a light microscope (VWR International, Inc.). A thorough microscopic analysis

was done, including examination of leaf structure, epithelial tissues, stem structure, hold fast structure, and overall health. Under the dissecting microscope, the plants were viewed in water to examine the original conditions. Epithelial tissues were carefully removed using a high-grade graphite scalpel, and then examined under a high-power light microscope. In addition, an analysis of salt sensitivity was done under a light microscope using a concentrated saline solution (Sigma-Aldrich Company). The stomata were also examined to determine damage and the effect of the calcium carbonate and ammonium chloride. The holdfast was examined under the dissecting microscope for evidence of damage to root hairs and overall color.

Microorganism Analysis

The water from each phase, including all jar tests and lacing procedures, was cultured onto blood/ MacConkey Agar plates (VWR International, Inc.) to test for various microorganisms. Samples of water from each phase were placed in sterile, vacuum-sealed test tubes (VWR International, Inc.). Six plates were made and labeled as distilled, control, storm drain 1, storm drain 2, storm drain 3, and agricultural runoff. For this phase, the distilled water streak was the control to test for purity of the plates. They were allowed to stabilize for 12 hours, and were then streaked using the standard three-step streaking procedure. Placement into a $35 \pm 1^\circ\text{C}$ incubator (Barnstead/Thermolyne, Conroe, Texas) was immediate. After growth for five days, the plates were removed from the incubator, and the present bacteria were mounted onto oil-microscopy slides (VWR International, Inc.). The common staining procedures (Juchau, 1995) were then followed. The slides were examined under an oil immersion microscope (VWR International, Inc.) for gram positive and negative cocci and bacilli.

The bacteria were further identified using two different approaches to ensure accuracy. The first approach involved culturing in different culture media (Barkocy-Gallagher et al., 2002). The bacteria were cultured onto blood (VWR International, Inc.), MacConkey (VWR International, Inc.), and Hektoen (VWR International, Inc.) agars to test for the presence of salmonella, shigella, virulent E. coli, and other enterobacteracea.

The second approach involved DNA isolation and gel electrophoresis. The DNA from the cultured bacteria was extracted using common phenol-chloroform lysis procedure with TRIzol (Invitrogen Corporation, Carlsbad, California) (Masri et al., 2002) and examined with gel electrophoresis (Owl Separation Systems, Portsmouth, New Hampshire).

Soil Sampling Procedure

Soil samples from various areas of Lake Machado and Madrona Marsh were taken for measurement of phosphate salt concentration, ammonium salt levels, pH, and consistency during the dry season. A 101-mm (4-in.) core of soil was removed using the LaMotte Soil Sampler and dried for four days in a hermetic container. The soil was then placed in distilled water, where various minerals leached into the water for measurement. In addition, a dry soil analysis was done. This included reaction with 1-M hydrochloric acid solution to test for carbonates and sulfites. Then, the soil was flash baked over open flame for removal of carbonate and reacted again with hydrochloric acid for confirmation of the presence of sulfites. Sulfur dioxide gas was also identified. The soil sample was also reacted with 1-M sodium hydroxide solution to test for metal ions. In addition, a Lugol's and Brachinoff's solution was used to test for organic solutes. The soils were examined under a dissecting microscope and a light microscope for identification of the various soil horizons and porous nature of the sample.

Spring Loading Filter Model Approach

The spring loading filter seems to be a convenient filter for use in small systems with relatively low flow rates, in accord with Stoke's Law (Yamada, 2000). It consists of two spring loaders (Toro Company, Bloomington, Minnesota), the first containing calcium carbonate and the

second containing activated carbon (PetCo). At the base of each spring loader, a particular type of filter material (fiber medium or heavy duty paper filter) was added. In each test, varying forms of calcium carbonate as resolidified powder, purified powder (Sigma-Aldrich Company), or marble chips (Quickrete Companies, Atlanta, Georgia) were also used. The inflow consisted of a 9-ppm sodium phosphate solution, the outflow was tested for phosphate concentrations, and turbidity and time measurements were recorded.

PVC Pipe Filter Model Approach

The PVC pipe filter seems to be a convenient filter for larger flowrates and water with high particulate concentrations. It consists of calcium carbonate followed by activated carbon and filter in one container sided by two PVC pipes (Home Depot, Inc., Atlanta, Georgia). Again, in each test, varying forms filter material (fiber medium or heavy duty paper filter) and calcium carbonate as resolidified powder, purified powder, or marble chips were used. The inflow consisted of a 9-ppm sodium phosphate solution, the outflow was tested for phosphate concentrations, and turbidity and time measurements were recorded.

Galvanized Steel Pipe Filter Model Approach

The galvanized steel model is convenient because in can be fit directly into galvanized steel pipes (Home Depot, Inc.), and the filters can be changed from the top. It consists of two filters that fit into grooves that are drilled into the pipe, the first containing calcium carbonate and the second containing activated carbon. In each test, varying forms of a particular type of filter material fiber medium or heavy duty paper filter and calcium carbonate as resolidified powder, purified powder, or marble chips were used. The inflow consisted of a 9-ppm sodium phosphate solution, the outflow was tested for phosphate concentrations, and turbidity and time measurements were recorded.

Agitator-Clarifier Filter Model Approach

The agitator-clarifier filter is good for large flowrates and water with high particulate and granular matter. This filter is still in its building and testing stage, and, because of its more complex nature, it will take some time for completion. It is being directly implemented into Madrona Marsh preserve with the approval and funding of the City of Torrance and the Friends of Madrona Marsh Board of Directors.

Results and Discussion

The results were very favorable. The normal phosphate content of oligotrophic lakes is approximately 0.1 ppm (Keenan and Lowe, 2001), but the control showed that Harbor Lake had twice this concentration of phosphates and Madrona Marsh had at least 10 times this concentration of phosphates—in some places, up to 30 times this concentration. Eutrophic waters can generally be considered those which have high nutrient levels, large quantities of plants and algal matter, low biological diversity, different species from noneutrophic waters, low overall levels of dissolved oxygen, big differences between day and night time levels, and overall poor water quality. These two bodies have most, if not all, of the preceding qualifications and can therefore be classified as eutrophic.

In the Lake Machado analysis, it was determined that all of the storm drains and agricultural runoff had high levels of phosphates (Figure 2). The agricultural runoff had an especially high level of phosphate ions. The dissolved oxygen content was low in all test samples, but noticeably low in the agricultural runoff. The fact that the pH in storm drain 3 was 7.6 compared to a 7.2 in the control signifies that storm drain 3 is 40% more basic than the lake water, which can be attributed to the various dissolved basic salts in the runoff, which was further resolved in the next phase. In the agricultural runoff, the pH was slightly acidic because of the fertilizer's

acidic salt content. The temperature increased slightly as a result of the small quantity of water and the presence of moderate direct sunlight.

Water collected from the indicated sources in Lake Machado in the first phase shows natural biological phosphate reduction rates in the lake without the addition of a chemical phosphate binder material. The control indicates the relatively stable phosphate conditions that exist during a relatively long period of time (approximately 3 weeks) without additional phosphate from any source. This indication suggests that phosphate management cannot be left to self-mitigation after removal of phosphates, but rather that active chemical intervention may be needed to ensure proper phosphate levels.

In phase two, calcium carbonate was tested as a potential phosphate binder in an effort to decrease phosphate concentration in the water. The calcium carbonate was highly successful ($p < 0.0001$) in decreasing phosphates from 1.5 ppm, on average, to 0.4 ppm (Figure 3), an average decrease of approximately 70%. The carbonate did raise the pH of the water slightly, but this effect was determined to be environmentally innocuous. It raised a slightly acidic pH to a pH very close to that of the control in phase 1 (7.2), quite possibly the best level of incoming water. In addition, no adverse effect on plant life was noticed. The dissolved oxygen content of the water was increased significantly ($p < 0.01$) (Figure 4) by the plant and the increase of carbon dioxide in the water. The ammonium chloride, while successful in decreasing the temperature slightly, did not aid in either of the purposes of decreasing phosphate by dissolving calcium carbonate or increasing dissolution of oxygen. It also increased the ammonium content of the water, the most probable cause for the poor condition of the plant exposed to this water. The temperature rose steadily in all of the samples with the salinity, indicating a certain level of evaporation and the effects to their exposure to room temperature. The calcium concentration dropped notably ($p < 0.0001$) between days 2 and 4, indicating that precipitation of calcium phosphate occurred. The results from this phase indicate that calcium carbonate is both a feasible and environmentally advantageous phosphate binder and that it holds the promise of a better fresh water ecosystem.

Currently, Madrona Marsh maybe considered highly polluted (Table 1). Sump water and water throughout the marsh, especially in the northwest corner, have average phosphate concentration of 2.34 ppm (7.51×10^{-6} mol/L). The dissolved oxygen content registers at 0 ppm (0 mg/L) in certain areas of the marsh, including the northwest corner. These levels cannot sustain significant native life in any form.

The amount of calcium carbonate needed to decrease phosphate, as determined from the reaction calculation, by a certain amount in a certain amount of water was calculated to be 4.78×10^{-4} moles per liter per ppm phosphate, as follows:

$$(\text{CaCO}_3) = K_{cp} \left([\text{PO}_4^{3-}] \right) (V), \quad (4)$$

where $K_{cp} = 4.78 \times 10^{-4}$ moles per liter per ppm phosphate For CaCO_3 in milligrams, $K_{cp} = 48.7$ The data from the calcium carbonate reaction mechanism analysis lead directly to the calcium carbonate lacing procedure. A phosphate level of 0.75 ppm was desired for testing purposes, and the results were very accurate. The average phosphate levels after the addition of calcium carbonate came to a statistically significant ($p < 0.001$) 0.74 ppm (Figure 5), which is well within the statistically significant range ($p < 0.01$). The average phosphate levels after the addition of calcium carbonate and concentrated hydrochloric acid came to a slightly lower average of approximately 0.68 ppm. Overall, this phase corroborated the data from the K_{cp} calculations. The pH was not affected by the addition of either the calcium carbonate or the hydrochloric acid, and no evidence of any damage to the ecosystem was found in the month-

long study. The levels of other ions were relatively stable, indicating that no significant extraneous reactions occurred. Dissolved oxygen levels rose in some of the test samples (Figure 6), corroborating the hypothesis that dissolved oxygen levels and phosphate levels are inversely related.

The ammonia reduction mechanism tests also showed significant results ($p < 0.01$) (Figure 7). There was no notable change in the ammonia concentrations in the control phase. The data showed, in some cases, a 6.7% decrease in ammonia within the first five days. Contrary to the hypothesis, it seems that aeration is more beneficial in terms of ammonia reduction than the fiber medium. This could be the result of several factors, including the fact that dissolved oxygen in the water was very low. The aeration increased the oxygen levels significantly ($p < 0.001$), even counteracting any negative effects of temperature increase. *Nitrosomonas* are especially sensitive to low oxygen levels, and this may have increased its growth. Even minimal increases in dissolved oxygen levels seem to increase their growth.

The presence of the following pathogenic bacteria was determined in the waters: *Escherichia coli* 0.57-H7, *Klebsiella*, *Serratia*, *Proteus*, *Citrobacter*, *Pseudomonas*, *Aeromonas*, and *Salmonella* (Table 2). The presence of these bacteria in a naturally oligotrophic lake indicates that the process of eutrophication has continued despite work from many environmental organizations. As shown by preliminary data, one possibility of decreasing these bacterial populations in this water is to decrease nutrient input and nutrient concentrations in the water (Figure 8).

Various filter models were tested as a major part of this project. The spring loading filter design was found to be the most effective in reducing the phosphates; however, it had the lowest flowrate of the three filters. The PVC filter design and the metal filter design both seemed to be equal in their performance. They did not have results as favorable as the spring loading filter, but their flowrates were more realistic (Table 3). The acrylic agitator-clarifier system is still in the building process. This is the next stage of the experiment. The City of Torrance is willing to have this system, fully tested, installed into the Madrona Marsh sump area as a purifier for the water that passes from the sump to the marsh proper. Part of this plan also involves the diversion of all storm drains to the sump before any water enters the marsh proper.

Conclusions

Both the lacing method and the filtration method of phosphate control seem to be very promising. The use of aeration and growth media is effective in reducing ammonia. Therefore, by using these two systems, eutrophication can be controlled, reducing phosphates, increasing dissolved oxygen levels, returning pH to a more natural level, and decreasing ammonia and nitrates through denitrification, while reducing algal and bacterial populations and, thus, increasing visibility and light penetration and decreasing turbidity in the water. Depending on the state of the individual lake ecosystem, one of the two of the options for the control of phosphates and ammonia must be chosen, or they can be used in conjunction. The imminent goal of the study is the implementation and testing of the developed filter at Madrona Marsh. These plans have been approved by the City of Torrance and are currently underway, and sufficient funding is being appropriated for this purpose. If this filter is successful in Madrona Marsh, as the evidence from this study indicates, plans for its implementation in Harbor Lake Machado and other local bodies of water will be considered.

Acknowledgments

Credits. My parents, Srinivas and Sita Yanamadala, gave me tremendous assistance and advice in the completion of this project, and it was their support that made this work possible. Peter Starodub and Mariana McMullen were critical in directing this research and served as my mentors and advisors throughout the three years during which I conducted

this work. Additionally, Jess Morton of the Palos Verdes/South Bay Audubon Society (California) provided invaluable advice. The City of Torrance (California) and the Friends of Madrona Marsh (Torrance, California) made available facilities for the conduction of these studies.

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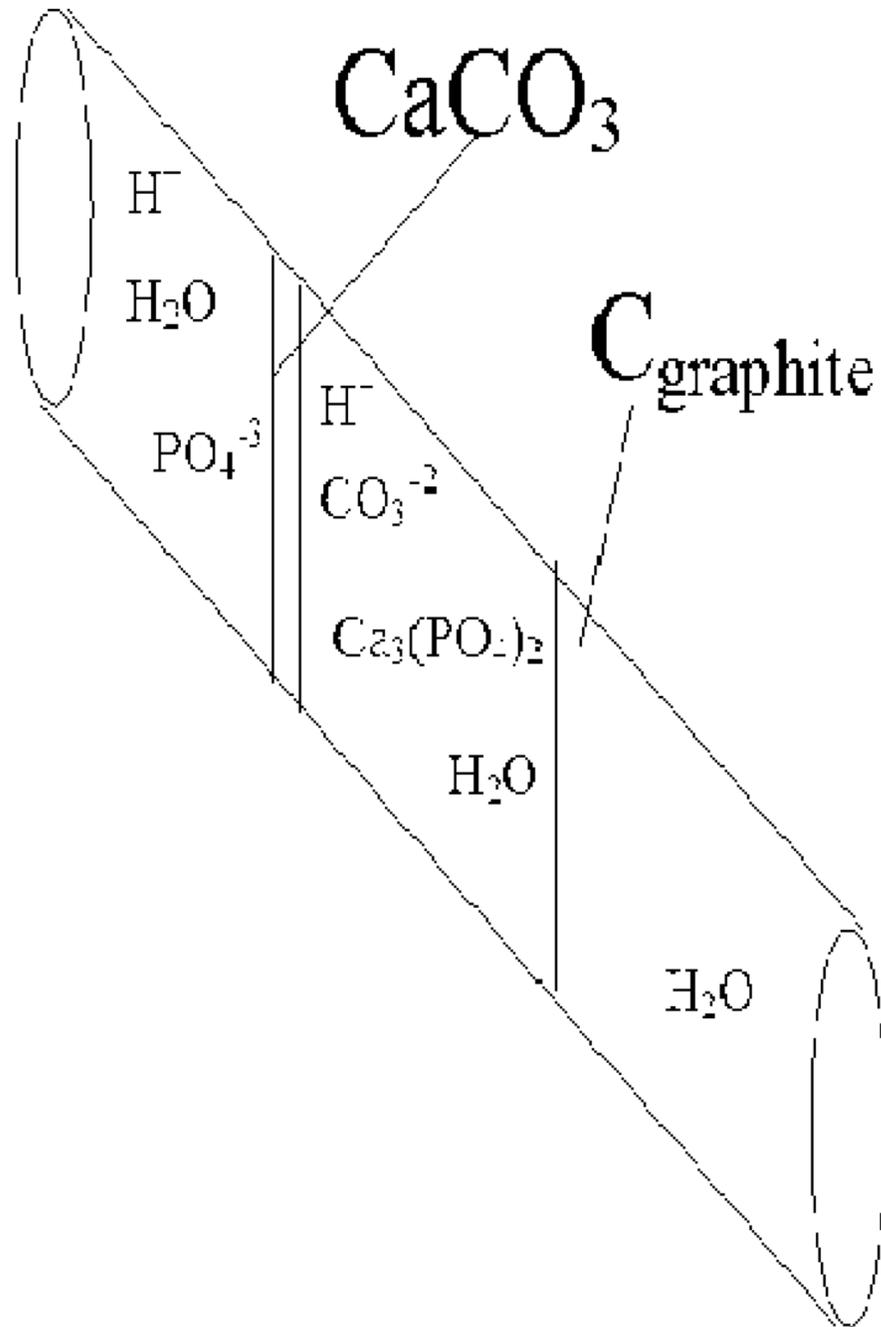


Figure 1.
Basic filter model design.

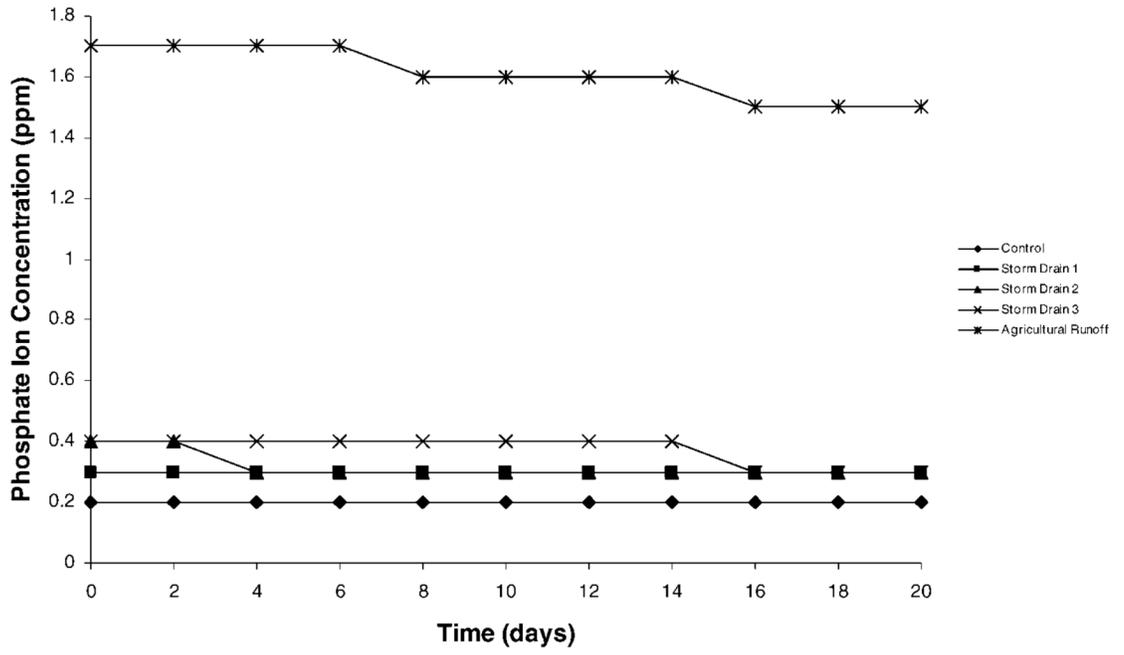


Figure 2.
Lake Machado phosphate concentrations.

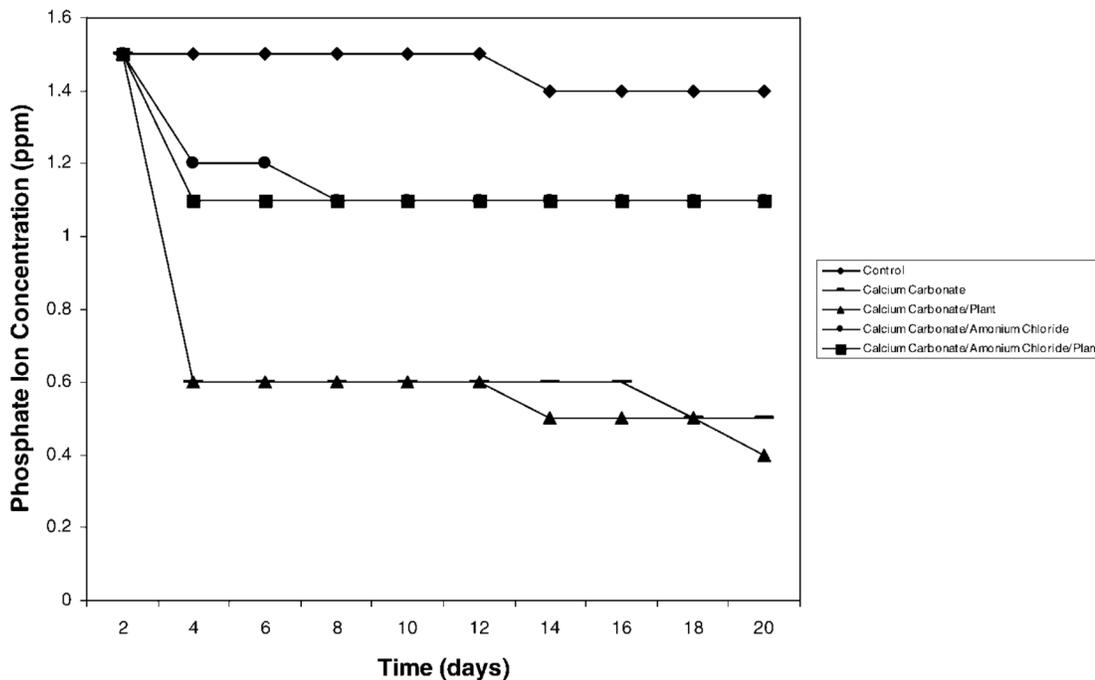


Figure 3. Calcium carbonate phosphate reduction in Lake Machado water. The water sample, taken from the area that had relatively stable three-week, shows significant ($p < 0.0001$) reduction in phosphate by calcium carbonate binding. Ammonium chloride had no increasing effect on calcium carbonate phosphate reduction.

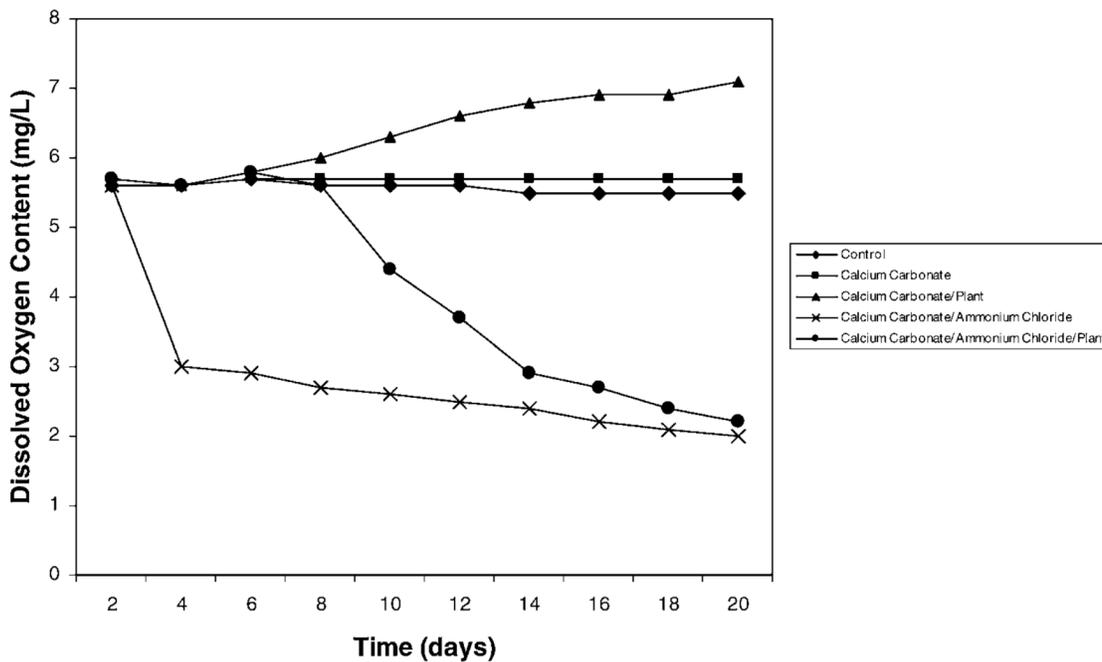


Figure 4. Calcium carbonate phosphate reduction resulting in significant ($p < 0.001$) increase in dissolved oxygen. Phosphate reduction by calcium carbonate shows a marked increase in dissolved oxygen alone and especially in conjunction with plant. The use of ammonium chloride, however, shows detrimental effects on dissolved oxygen concentrations, leading to more anoxic conditions.

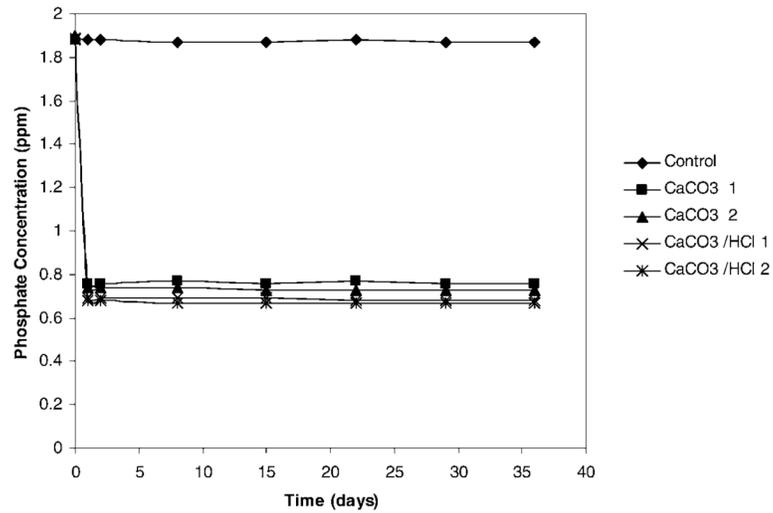


Figure 5. Calcium carbonate lacing procedure in Madrona Marsh. Lacing carried out on site at Madrona Marsh shows the efficacy of calcium carbonate phosphate reduction in the actual ecosystem and the accuracy of the calcium carbonate formula, which predicted final phosphate concentrations of 0.70 ppm.

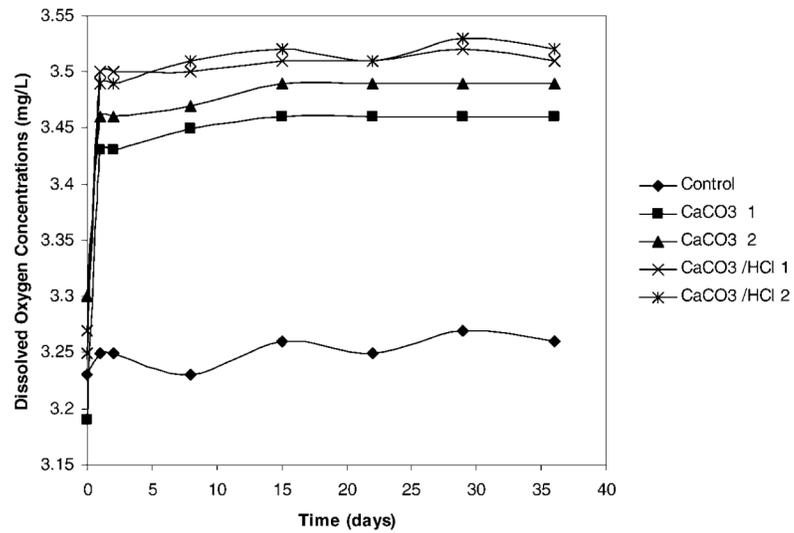


Figure 6. Dissolved oxygen increases in response to calcium carbonate phosphate reduction. Significant ($p < 0.0001$) increases in dissolved oxygen are seen repeatedly with calcium carbonate phosphate reduction, as seen here in the lacing procedure conducted within Madrona Marsh.

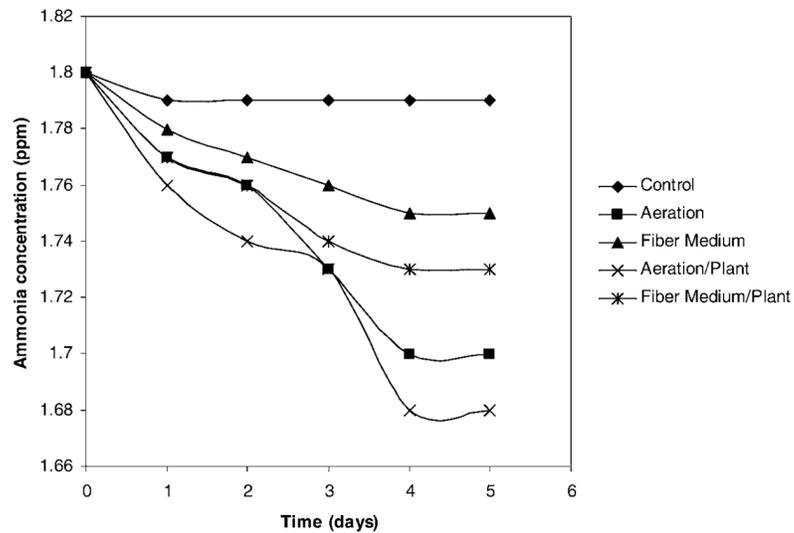


Figure 7. Ammonia reduction. Ammonia reduction through aeration, the use of a fiber medium for increased growth of *Nitrobacter* and *Nitrosomonas*, and the use of plants all show significant ($p < 0.01$) decreases in ammonia within a 5-day period. Ammonia reduces the efficiency of calcium carbonate phosphate reduction, and reducing ammonia concentrations has shown to restore this efficiency.

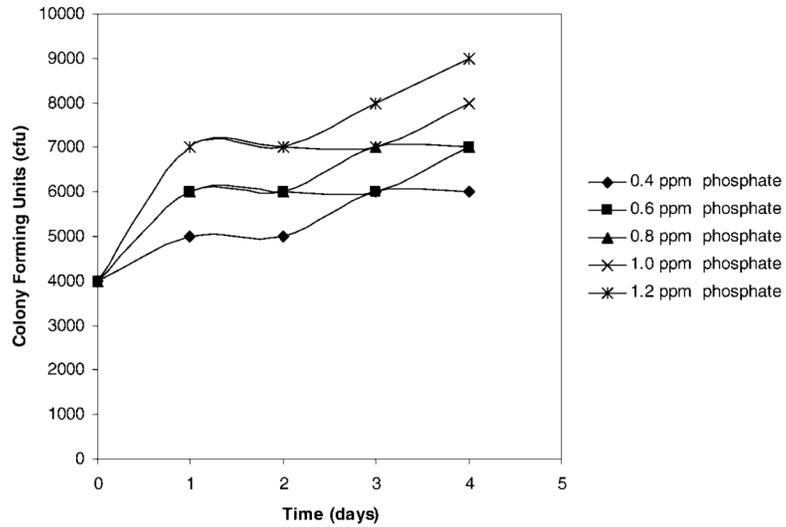


Figure 8. Effects of phosphate reduction on potentially pathogenic bacteria. A 66% reduction in phosphates shows a 35% reduction in potentially pathogenic bacteria as cultured on Hektoen Agar. Further analysis will be conducted to corroborate this finding.

Madrona Marsh analysis. Madrona Marsh water taken from 10 sources within the marsh show significant phosphate and nitrate pollution and very low dissolved oxygen concentrations. Levels of other pollution indicators are also above normal levels.

Table 1

ppm	PO ₄ ³⁻	NO ₃ ⁻	O ₂	NH ₃	Cl ⁻	Fe	Cu	Cr ⁶⁺	pH
1	1.97	13.97	3.06	0.55	0.1	0.23	0.08	0.01	7.3
2	4.31	14.07	0.31	0.47	0.9	0.57	0.01	0.02	7.7
3	4.57	14.16	0.29	0.39	0.8	0.49	0.02	0.01	7.7
4	2.09	13.39	2.39	0.43	0.3	0.24	0.01	0.05	7.4
5	1.99	12.01	3.54	0.57	0.5	0.17	0.02	0.03	7.3
6	1.73	11.17	5.36	0.61	0.1	0.11	0.03	0.04	7.2
7	1.99	13.17	2.86	0.54	0.3	0.17	0.00	0.03	7.3
8	1.99	13.55	1.96	0.54	0.1	0.19	0.02	0.04	7.4
9	1.97	13.47	3.86	0.57	0.3	0.12	0.12	0.12	7.3
10	1.96	12.72	4.01	0.55	0.2	0.11	0.11	0.11	7.3

Table 2

Bacterial culture analysis. These bacteria have been found within Madrona Marsh, causing alarm in terms of public health and ecosystem management.

MacConkey Agar

Escherichia coli (nonpathogenic)

Kelbsiella (pathogenic)

Serratia (pathogenic)

Proteus (pathogenic)

Citrobacter (pathogenic)

Escherichia coli (pathogenic)

Pseudomonas (pathogenic)

Aeromonas (pathogenic)

Hektoen Agar

Salmonella (pathogenic)

Table 3

Calcium carbonate phosphate reduction in filtration. Calcium carboante significantly ($p < 0.0001$) reduced phosphate concentrations from 9.0 ppm to the given values in quick filtration through the three models. This type of filtration system is currently under planning and construction at Madrona Marsh preserve in Los Angeles County, California.

Filter model	Filter material	Filter medium	Phosphate (ppm)	Turbidity (NTU)	Time (s)
1 (spring)	Fiber medium	CaCO ₃ powder	0.58	4	10
		Marble chips	0.41	5	10
		Solidified CaCO ₃	0.22	7	12
2 (PVC)	Standard grade	CaCO ₃ powder	0.49	2	13
		Marble chips	0.39	2	14
	Fiber media	Solidified CaCO ₃	0.17	3	18
		CaCO ₃ powder	0.59	4	7
3 (galvanized)	Standard grade	Marble chips	0.43	6	8
		Solidified CaCO ₃	0.24	8	10
		CaCO ₃ powder	0.51	2	9
	Fiber medium	Marble chips	0.42	3	11
		Solidified CaCO ₃	0.19	5	14
		CaCO ₃ powder	0.58	4	9
Standard grade	Fiber medium	Marble chips	0.42	5	11
		Solidified CaCO ₃	0.21	8	12
	Standard grade	CaCO ₃ powder	0.51	2	11
		Marble chips	0.39	3	13
		Solidified CaCO ₃	0.18	4	15