SURVEY OF THE WATER CHEMISTRY AND SPECIES DIVERSITY OF VERNAL PONDS IN NORTHERN MICHIGAN

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ABSTRACT.- This survey was conducted to study the water chemistry and species diversity on twenty-seven vernal ponds in Gogebic County, Michigan at the University of Notre Dame Environmental Research Center, UNDERC. Vernal ponds are often separated into groups based on their great degree of biological and physico-chemical variation (Pusey 1990). Great variation in the chemical composition of the vernal ponds was observed in this survey. The data collected was analyzed in relationship to size, location on the property, chemical changes within the ponds, and the organisms present. Tests were performed in the field and in the laboratory. Nalgene bottles were used to collect the water and then frozen for later laboratory testing. Organisms were preserved in 70% ethanol for later identification. The samples were then analyzed in the laboratory for different properties and chemical species. There was a direct correlation found between pond area and alkalinity. In addition, species diversity was directly correlated with hardness but indirectly related to aluminum concentration. A definite relationship was found between species diversity and pond maximum depth and area. Larger, deeper ponds generally had a greater species diversity. This study serves as the foundation to analyze the small community of vernal pond biological activity as a whole. There may also be other factors besides chemistry needing further in depth analysis such as; size, depth, benthic layer material, light (canopy), etc.-Baskin 1994.

INTRODUCTION

By surveying the water chemistry of vernal ponds, one is able to gain information concerning the organismal composition of local aquatic communities. Vernal ponds result from the accumulation of water from the annual snowmelt and spring rains in shallow depressions and become dry by the mid to late Summer (Rowe 1993). The different compounds and elements of interest provide a means to study the important relationship between the physical and chemical properties and the fauna in the ponds. The study was conducted on twenty-seven vernal ponds in Gogebic County, Michigan at the University of Notre Dame Environmental Research Center in the summer of 1995. The altitude of the property ranges from 500 meters to 520 meters above sea level. The vernal ponds are distributed over a land area of 2485 ha.

Previous studies on the effects of chemical parameters on organisms that inhabit vernal ponds fail to provide a full understanding of the relationship. The primary organisms that have been studied are salamanders. One study focused primarily on the relationship between the pH of the water and the reproductive and developmental success of three different species of larval amphibians, *Ambystoma jeffersonianum*, *Ambystoma maculatum*, and *Rana sylvatica* (Rowe 1992). The experimental results showed that at pH levels of 4.2, the mortality of *A. jeffersonianum* increased (Rowe 1992). In addition, the mean wet masses of *R. sylvatica* were found to decrease and the time to metamorphosis increased (Rowe 1992). The leaching of Aluminum from the sediments into the water column was also thought to affect the amphibians. In another study, the pH of vernal ponds was found to affect the reproductive success of Jefferson salamander adults (Horne 1994).

The purpose of this survey is to evaluate the relationship of the water chemistry data, relative pond areas, and the species that inhabit each vernal pond. The species survey was comprehensive and included the identification of insects, arachnids, arthropods, molluscs, and amphibians. No fish were expected to be found in the pools. The survey should be able to give distribution patterns of the species present in the pools. One will then be able to compare this data to other information about the pools such as the water chemistry and perhaps be able to learn the reasons for the distribution patterns. In addition, one also uses the information gathered to discover possible harmful effects of the chemical elements and compounds on the organisms that live there as well as the effects that the organisms may have on the water chemistry.

Furthermore, this study will provide data that can be referred to as the years progress. In recent years, a world-wide decrease in the amphibian population has been documented. Due to the lack of fish predation, amphibians use the ephemeral waters to breed, and they are a major component of the fauna in the vernal ponds. Although many theories have been presented, previous data on the locations is often unavailable for comparison. However, when a noticeable change in species composition (including amphibians) occurs in any of the vernal ponds surveyed, the water chemistry can be compared to this previous study to see if the chemistry may be the cause. In addition, the land where the study is taking place is free from anthropogenic influences. Therefore, the vernal ponds can be studied for natural fluctuations occurring in both the chemical and biological environments. In conclusion, water chemistry can be used as a tool in describing the distribution of the many organisms that rely on the vernal ponds as breeding sites.

MATERIALS AND METHODS

The study was conducted on twenty-seven different roadside vernal ponds presently studied by Dr. George B. Craig, Jr. on the property (Figure 1). The chosen sites were surveyed May 26-27, 1995 for water chemistry, and all of the specimens were collected on May 24, May 27, June 2, and June 3, 1995. At the site, the length, a few horizontal parameters, and maximum depth were measured using tape measures. The horizontal parameters were taken to give an outline of the pond's general shape. The measurements were taken after the chemical data and samples were collected to prevent disturbance.

The following tests were done in the field; temperature, dissolved oxygen, conductivity, total dissolved solids, and pH. First of all, an oxygen/temperature meter and probe was used to measure the air and water temperature and oxygen saturation. A conductivity meter was used to measure both conductivity and total dissolved solids. The pH was also measured using a pH meter. The water readings were taken at approximately 10 cm from the pond's surface with each probe. Two 250 ml Nalgene bottles were used to collect water samples, without any air, from each pond. The two water bottles, collected at the same place and at approximately the same depth, were then frozen for testing at a later date.

Once the samples were thawed to room temperature, they were tested for their chemical content. All tests were conducted in previous rinsed glassware. The Hach Chemical Analysis Kit was used for the chemical tests (Hach Chemical Co., 1975). The tests for alkalinity and total hardness were performed by using the method of titration and reagents from the Hach kit. Such a method has a high error if not performed with precision. The other tests were conducted by adding different reagents to the pond water samples and then were read by the spectrophotometer. The reagents added to the samples of pond water were in premeasured packets. Therefore, error in adding the specified amount of the chemicals was negligible.

In the laboratory, the following eleven tests were conducted on each sample; Nitrogen,Ammonia-Nessler Method (0-2.5 mg/L NH_4/NH_3), Nitrate,Low Range-Cadmium Reduction Method (0-0.4 mg/L NO_3/NO_2), Phosphorus,Reactive-Ascorbic Acid Method (0-2.5 mg/L P reactive/total), Sulfide-Methylene Blue Method (0-0.6 mg/L H_2S), Sulfate-SulaVer 4 Method (0-70 mg/L SO_4/SO_3), Iron,Total-TPTZ Method (0-1.8 mg/L Fe), Aluminum-Aluminon Method (0-0.8 mg/L Al^{3+}), Tannin and Lignin-Tyrosine Method (0-9.0

mg/L), Color,True and Apparent-APHA Platinum-Cobalt Standard Method (0-500 units PtCo), Alkalinity-Buret Titration Method, and Hardness,Total-ManVer2 Buret Titration Method. The tests for the following chemical species depended upon the accuracy and precision of the spectrophotometer. The errors for the tests are as follows; Ammonia- ±0.015 mg/L NH_3, Nitrate- ±0.010 mg/L NO_3, Phosphorus- ±0.01 mg/L P, Sulfide- ±0.003 mg/L H_2S, Sulfate- ±0.9 mg/L SO_4, Iron- ±0.009 mg/L Fe, Aluminum- ±0.016 mg/L Al^{3+}, and Tannin- ±0.08 mg/L. The error for both the alkalinity and hardness tests is ±0.1 ml.

The species survey was non-quantitative. The samples were collected by hand, dip nets, and delta nets. The collection was done for thirty minutes by two people. The insects, arachnids, arthropods, and molluscs were all preserved in 70% ethanol in plastic bags for later analysis. The amphibians were first euthanized with 2% benzocaine and then placed in 70% ethanol to preserve them for later identification. Only one species of each amphibian was preserved for the entire survey. Additional amphibians collected were identified and released.

Once all the samples were collected and preserved in plastic bags, the sorting process began. Once identified, the specimens were placed in vials for long term storage. A reference collection of one of each species found was kept for each vernal pool (except for the amphibians). The insects were identified using <u>Aquatic Insects of Northern Wisconsin</u> and <u>An Introduction to the Aquatic Insects of North America</u>. The arachnids were identified with <u>How to Know the Spider</u>.

Regression analysis was performed on the chemical tests against both the pond area and the number of species found of each vernal pond. The statistical program utilized was SYSTAT, INC (VBL 5.04 for Windows).

In addition, descriptive statistics were collected for this experiment because all of the results simply describe the vernal ponds on that particular day and do not provide concrete information from which to use as model data. Since no replicates were done, accuracy may be of significant error. Precision allows for how close the results are within the given test errors of the spectrophotometer. Bias can result in errors of many different kinds. The errors may be due to the person conducting the experiments. For example, how the person decides which is the exact point where the solution turns red or blue in the titrations may have a great effect on the results of the hardness and alkalinity tests. Random errors may be due to the contamination of the samples by air or unclean glassware. Another cause of error may be improper reading of the meters and use of the probes.

RESULTS AND DISCUSSION

Chemical analysis did illustrate a wide variation between the different ponds. The amount of ammonia varied from 0.5 mg/L to the maximum value of 2.5 mg/L. The pH values ranged from 5.2 - 8.3. Al^{3+} was found to be in the range of 0.01 mg/L- 0.13

mg/L. The percent saturation of oxygen ranged from the undersaturated value of 30% to 100% saturation. Color of the water samples ranged from clear water with a value of 0.0 to the darkly stained water of 396 units PtCo.

The regression analysis found five significant trends. The trends are the following: 1) species number versus pond area, 2) species number versus maximum depth, 3) alkalinity versus pond area, 4) species number versus hardness, and 5) species number versus aluminum. As indicated, the only significant chemical test against pond area was alkalinity. In addition, Figures 6 - 21 illustrate the relationships of other variables. However, these were not found to be significant and are only included for future comparisons.

The regression test of species number versus pond area revealed a significant relationship (n=27, R^2=0.322, P=0.0017). The larger the area of the pond the more diversity of species were likely to be present (Tables 1 and 2). 32.2% of the species variance was due to pond area. One possible explanation is that a larger pond presents a greater likeliness of a diverse habitat. Larger ponds are more likely to have a tree fall into them which would create shelter and food. Larger ponds are also more likely to have grass growing in parts of them and no grass in other parts. This adds diversity of food sources and shelter. Larger ponds can also support more predators. There are more plant eaters present for the predators to hunt. However, one can not assume that pond area is the only variable affecting species diversity. Well-shaded pools may support less diversity than well-lit pools. Smaller pools are more easily shaded than bigger ones. Shaded pools may have smaller algae populations which would reduce a major food source for some insects. The aluminum and hardness of the ponds also play a role as related later in this paper.

Another important factor in species diversity was the maximum depth of a pond (n=27, R^2=.194, P=0.021). It seems the 19.4% of the variability in species diversity is dependent on the maximum depth of each pond (Tables 1 and 2). The major reason for this is probably a matter of habitat diversity. The greater depths allow for darker and cooler habitats. This may allow species to hide from their predators. Although the maximum depth is important in species diversity, it is clear that there are other factors involved. The factors mentioned before include hardness, aluminum content, and surface area. Light could once again play a role for the reasons explained earlier. Less light means that there is less algae to feed the higher organisms.

In addition, the regression analysis of alkalinity versus pond area illustrated a significant trend (n=27, R^2=.140, P=.055). The larger the pond area the greater the alkalinity value was (Figure 2). 14% of the variation in the data was due to alkalinity. Alkalinity is the measurement of the amount of carbon dioxide present in the water. The direct correlation between pond area and alkalinity may be related to the increased amount of species in ponds of greater pond area as previously discussed. Consequently, the greater the pond area and the number of species, the greater the respiration would be and therefore the amount of carbon dioxide. The amount of carbon dioxide would then increase due to the greater number of species, therefore increasng the alkalinity in these ponds. Although not thoroughly studied, the source of the water, for example, overland flow, may add to the alkalinity values as well as possible nutrient rich soil. In addition, alkalinity has been found to decrease due to the decrease of dissolved oxygen by the process of photosynthesis. Therefore, canopy cover may be another contributing factor. The lack of canopy allows more sunlight and consequently aids photosynthesis, causing a decrease in alkalinity. Further study of the effects of these three variables, overland flow, soils, and canopy density, on this relationship may prove to be important.

Furthermore, the relationship between the species number and hardness was shown to be significant. The regression analysis supports this apparent trend ($n=27$, $R^2=.157$, $P=.021$). From this data, 16% of the variation is due to hardness, which is the measure of bivalent cations. According to the statistical data, hardness was directly related to species richness (Figure 3). Since hardness includes calcium, magnesium, and other ions necessary for development, it seems intuitive that pools containing high amounts of these essential nutrients would also exhibit a large number of species. The increased hardness of the water seems to both support and nurture the various species found there. However, it is important to find from where the increases in the hardness content are coming. The source may be from surface flow or soil richness. Furthermore, a combination of other factors may play a small role in the increase of the species numbers, such as maximum depth.

Finally, another important trend was species richness versus the aluminum concentration. The regression analysis was also performed on this trend (n=27, R^2=.143, P=.043). As the concentration of aluminum increased, the number of species decreased (Figure 4). The data suggests that 14% of the variation is due to aluminum concentrations. This trend was perhaps the most predictable from the available literature. As the work of Rowe (1992) and Horne (1994) illustrates, increases in aluminum concentration often inhibit normal reproductive capabilities and development in amphibians. However, their work focused heavily on the relationship between the amphibians and the pH levels. Consequently, a trend was found between the pH levels and the aluminum concentration of the vernal ponds (Figure 5). As the aluminum concentration increased, the pH levels decreased. The work of Rowe and Horne illustrates that low pH levels also exhibiting higher aluminum concentrations corresponds with a decrease in the amphibian population. Although pH levels versus the species number was not significant, P=.538, the amount of aluminum was shown to have adverse affects. An explanation may be that soil was a major factor in the aluminum concentrations. As the pH levels dropped, soils containing significant amounts of aluminum leached the aluminum into the water, therefore affecting the species diversity.

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Figure 1 : University of Notre Dame Environmental Rearch Center (UNDERC) Gogebic County, Michigan

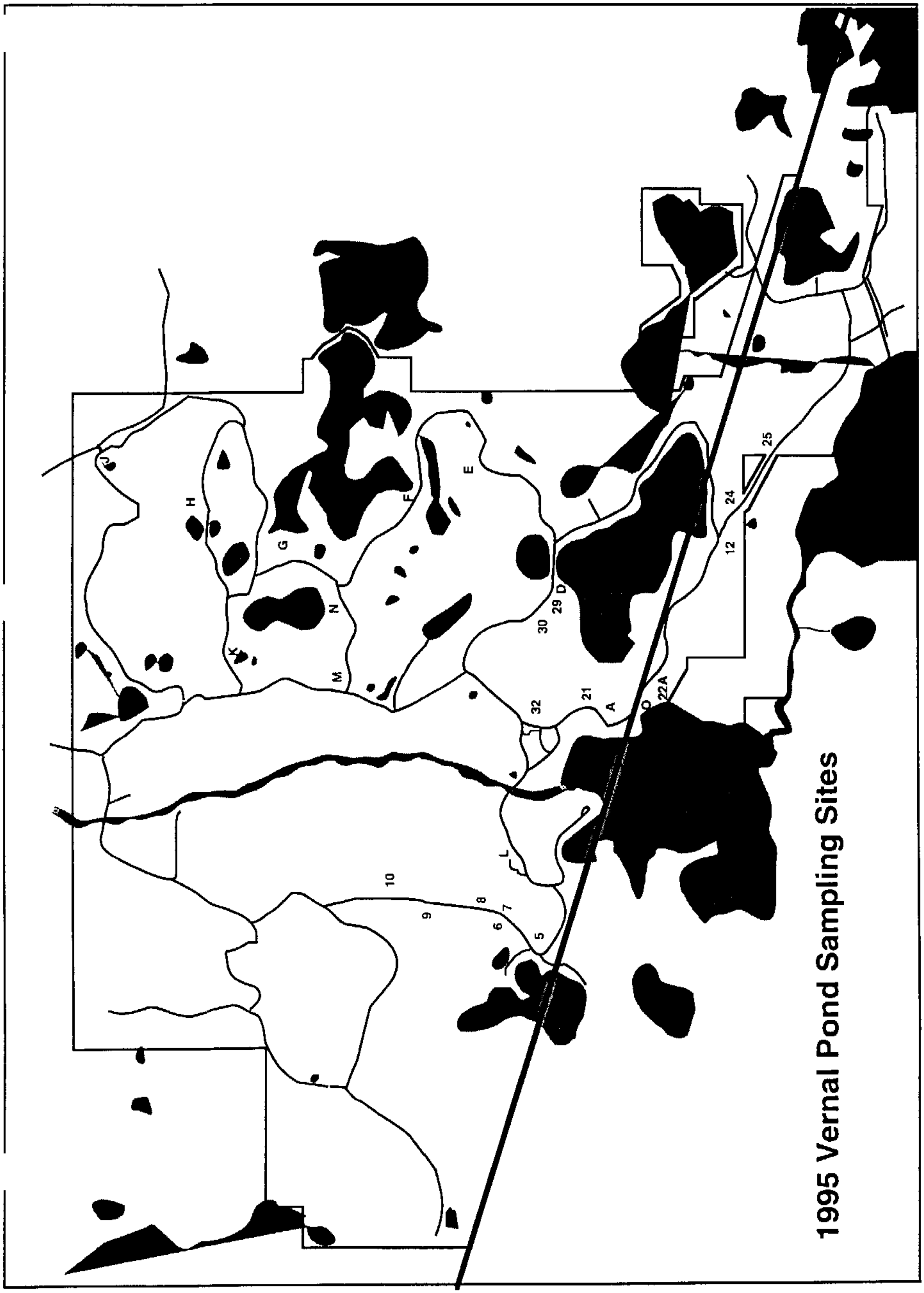


Table 1: The following table lists each vernal pond and the species present.

Site	# of Species	Species Present
5	10	*Rana catsbiena, Anostraka, Megadrilli, Agabus, Machlonyx, SHARIDAE, Acilius, Dixella, Matus, Laccophilus*
6	10	*Gerris, Sylvatica, Cordulogaster, SPHARIDAE, HAPLOTAXIS, Culex, Matus, Laccophilus*
7	4	*Gyrinus, Hyla crucifer, CULICIDAE, Eucorethru underwoodi*
8	7	*Rana sylvatica, Eucorethru underwoodi, SPHARIDAE, Aedes, Laccophilus, Gyrinus, CULCIDAE*
9	10	*Rana sylvatica, Bufo americanus, Dolomedes, Pirata, Physella, Gyrinus, CULICIDAE, SPHARIDAE, Limnophilus, RHANTUS*
10	10	*Dolomedes, Hespercorixa, Gyrinus, Eucorethra underoodi, Pusillus, SPHARIDAE, Limnophilus, Tropisternus, Laccophilus, Acilius*
21	8	*LUMBRICULIDAE, Trigoma, SPHARIDAE, Dixella, Gerris, Limnophilus, Dytiscus, ANURA (Tadpole)*
22	7	*SPHARIDAE, ANOSTRAKA, Dixella, Aedes, Dytiscus, Hespercorixa, Matus*
22a	10	*GASTROPODA, SPHARIDAE, ANOSTRAKA, Machlonyx, Aedes, Culicoides, Agabus, Dytiscus, CAUDATA (eggs)*
24	11	*Clubiona, Megadrilli, Fossaria, Mimetus, Gerrus, Gyrinus, Aedes, Limnophilus, Cymatia, Agabus, Rana sylvatica*
25	12	*Megadrilli, Physella, Dolomedes, Gerris, SHPARIDAE, Gyrinus, ANOSTRAKA, Lestidae, CULICIDAE (Pupae), Aedes, Columbetes, ANURA (Tadpole)*
29	8	*Gyrinus, SPHARIDAE, HAPLOTAXIS, Fossaria, Copelatus, Rana Clamitans, Ramphocorixa, Celmanthus*
30	11	*Limnophilus, Lenarchus, HIRUDIDIDAE, PHYSIDAE, Lestes, Aebetes acciductus, Hydactus, Acilius, Copelatus, Caudata (larvae), SPHARIDAE*
32	11	*Dolomedes, PHYSIDAE, CAUDATA (eggs), ANURA (eggs), HIRUDIDIDAE, Lestes, Gyrinus, Agabus, Eucorethra underwoodi, Acilius*
12a	14	*SPHARIDAE, PHYSIDAE, Dolomedes, ANOSTRAKA, Gyninus, Lmnophilus, Hespercorixa, Aedes, Ptychoptera, Laccophilus, Coleoptus, CAUDATA (eggs and larvae)*
A	10	*Gyrinus, SPHARIDAE, LUMBRICULIDAE, Dixella, Aedes, Agabus, HYdacticus, Oreodutes scitilus?, Dytiscus, Pericoma*
C	9	*Machlnyx, Dixella, Agabus, SPHARIDAE, Acilius, ANOSTRAKA, Megadrili, Culicoides, Limnohilus*
D	12	*ANOSTRAKA, Gyrinus, Heaspercorixa, Graphoderus, Mochlonyxx, OLIGOCHAETA, Progomphus obscurus, Laccophilus, Ilybus, Limnophilus, ANURA (tadpoles)*

E	6	*SPHARIDAE, Dolomedes, Haplotaxis, Agabus, Graphoderus, Limnophilus,*
G	3	*OLIGOCHAETA, Laccophilus, Limnophilus*
H	19	*Fossera, SPHARIDAE, Dolomedes, Hydryphontoidea, CAUDATA (tadpoles), Gerris, Rhamphocorixa acuminata, SALDIDAE, Agabetes acciductus, Oreodytes scitulus?, Graphoderus, Laccophilus, Aedesomatidae, ANNELIDA, Lestes, Culex, Limnophilus*
J	11	*SPHARIDAE, PIRATA, Gyrinus, Dixella, Lestes, Plothams lydia, Celithemis, Acilius, CAUDATA (tadpoles), ANURA (tadpoles)*
K	8	*Dolomedes, CULICIDAE (pupae), Gyrinus, Aedes, Hespercorixa, Laccophilus, Agabus, Rana pipiens*
L	12	*SPHARIDAE, Dolomedes, HIRUDINIDAE, Gyrinus, Gerrus, CULICIDAE (pupae), Hydactus, Hespercorixa, Laccophilus, Coleoptus, Ilybus*
M	11	*PIRATA, Haplotaxis, SPHARIDAE, CAUDATA (larvae), CULICIDAE (pupae), Gyrinus, Eucorethra underwoodi, Dixella, Lestes, Celithemis, Acilius.*
N	14	*PIRATA, CAUDATA (tadpole), Dolomedes, CLUBIONIDAE, Physella, SPHARIDAE, Gyrinus, Eucorethra underwoodi, Dixella, Graphoderus, Agabus, Limnophilus, Laccophilus, Rana catsbienna*
O	7	*Megarili, SPHARIDAE, ANOSTRAKA, Ilybus, Dixella, Hydroporus, CAUDATA (eggs)*

Table 2

Size Rank	Pond #	Area (m^2)	Size Rank	Pond #	Area (m^2)
1	m	22	15	32	104
2	7	25	16	l	105
3	5	29	17	a	117
4	k	30	18	22a	129
5	e	44	19	25	138
6	8	45	20	29	149
7	c	66	21	12	156
8	6	67	22	o	170
9	f	51	23	9	212
10	21	76	24	n	248
11	g	79	25	j	265
12	10	83	26	24	479
13	d	98	27	h	616
14	30	99			

The previous table lists each vernal pond by its area.

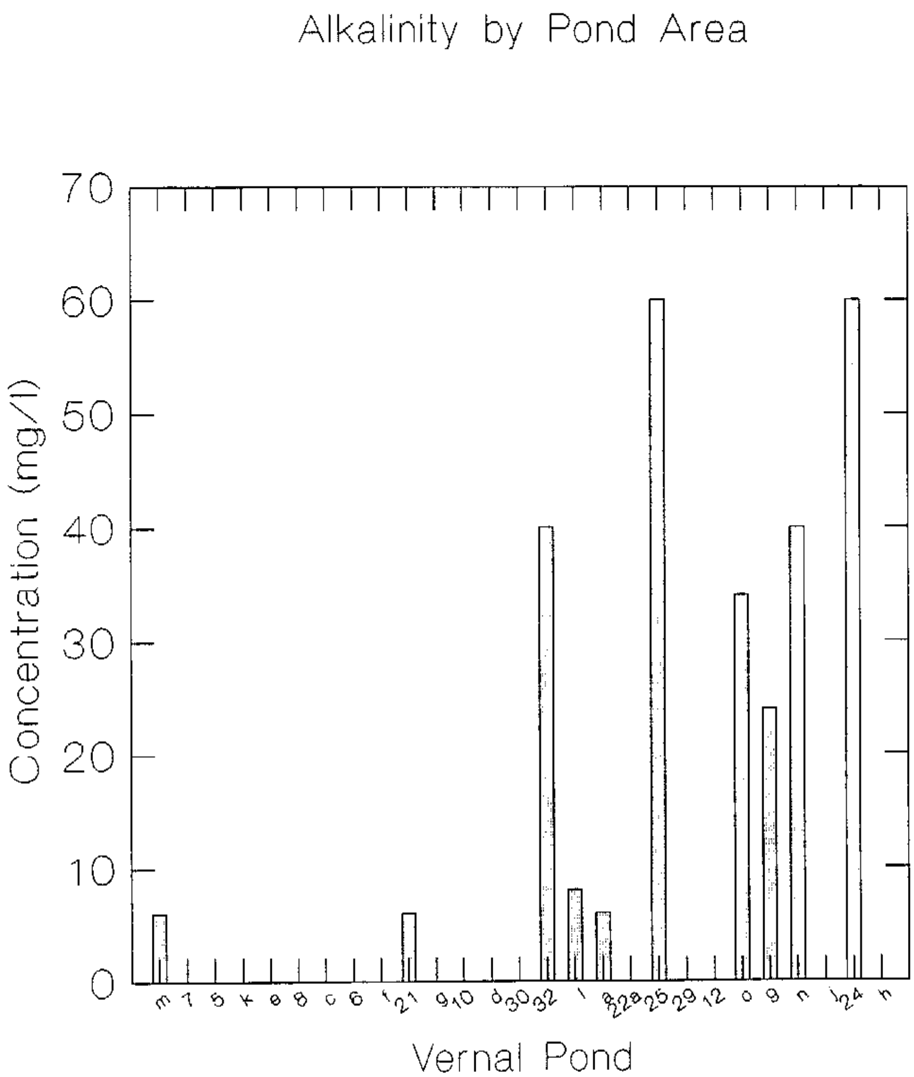


Figure 2: The graph illustrates the direct relationship between alkalinity and pond area.

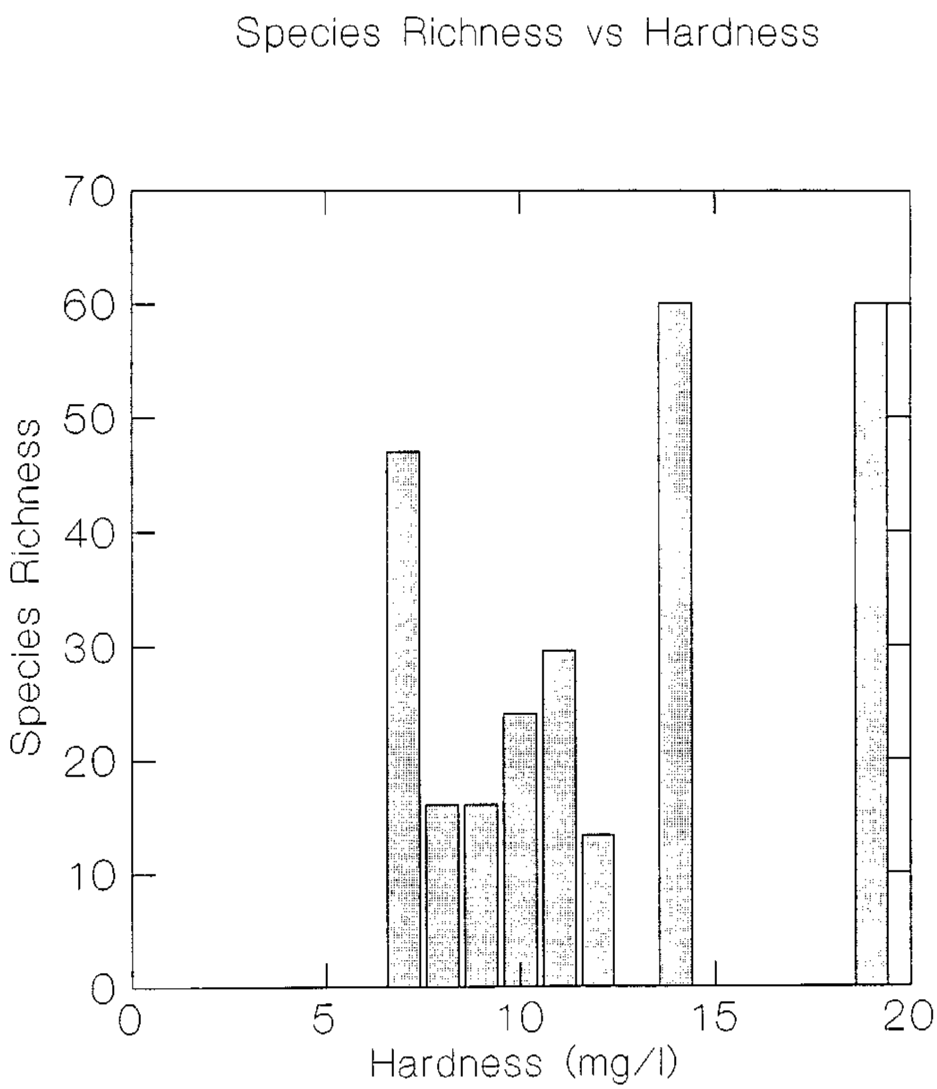


Figure 3: The graph illustrates the direct relationship between hardness and species richness.

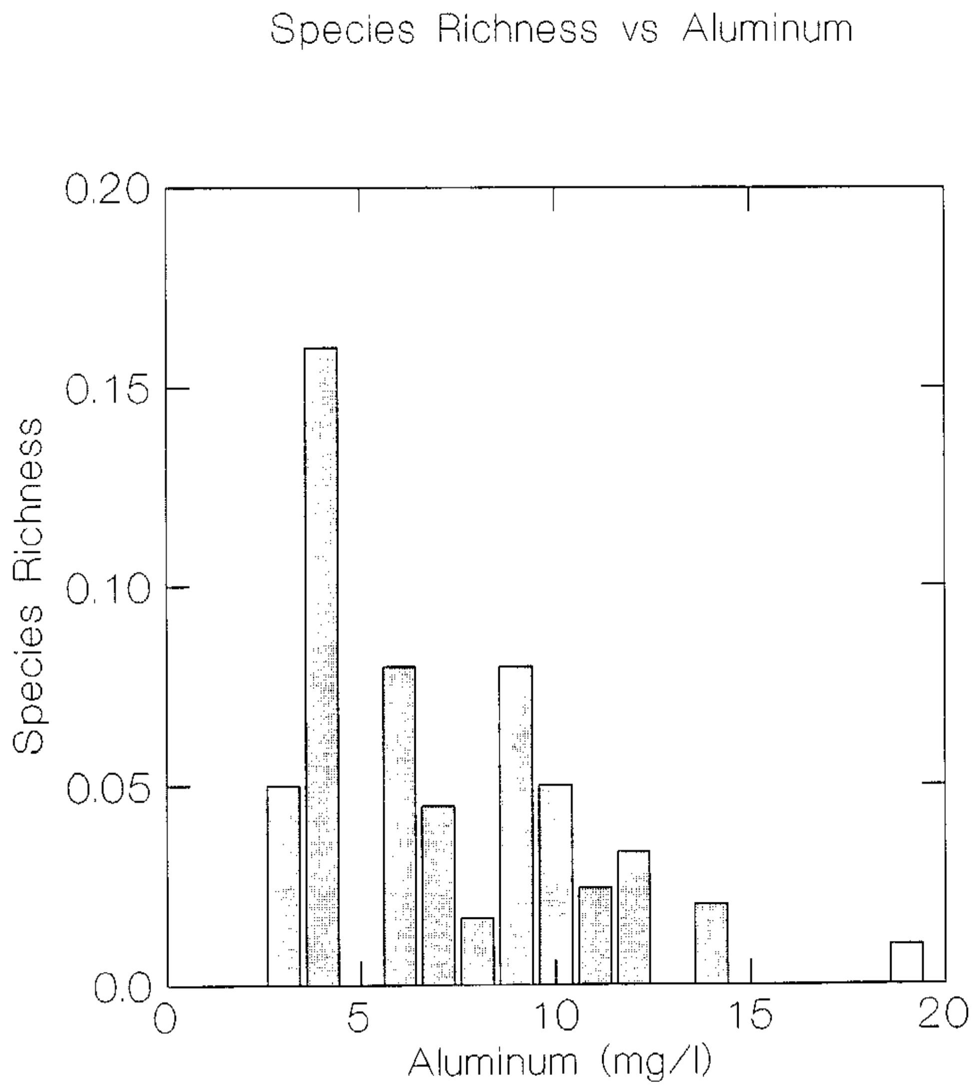


Figure 4: The graph illustrates the indirect relationship between aluminum and species richness.

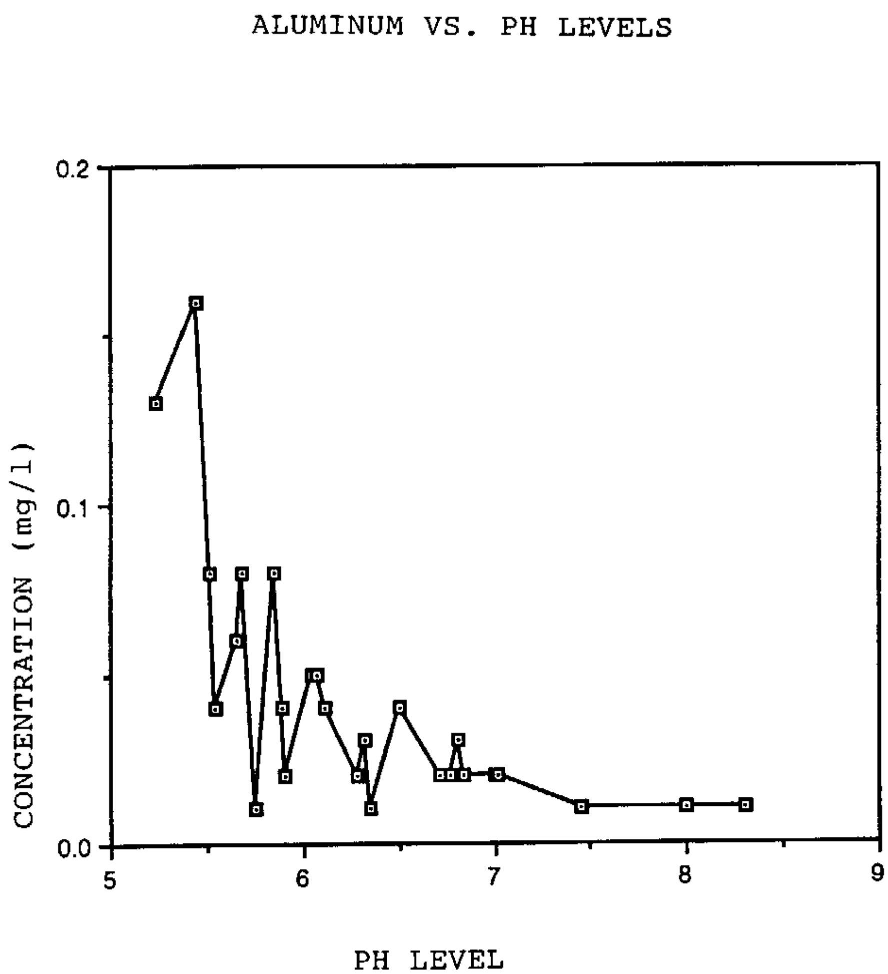


Figure 5: The graph illustrates the indirect relationship between aluminum concentration and pH levels.

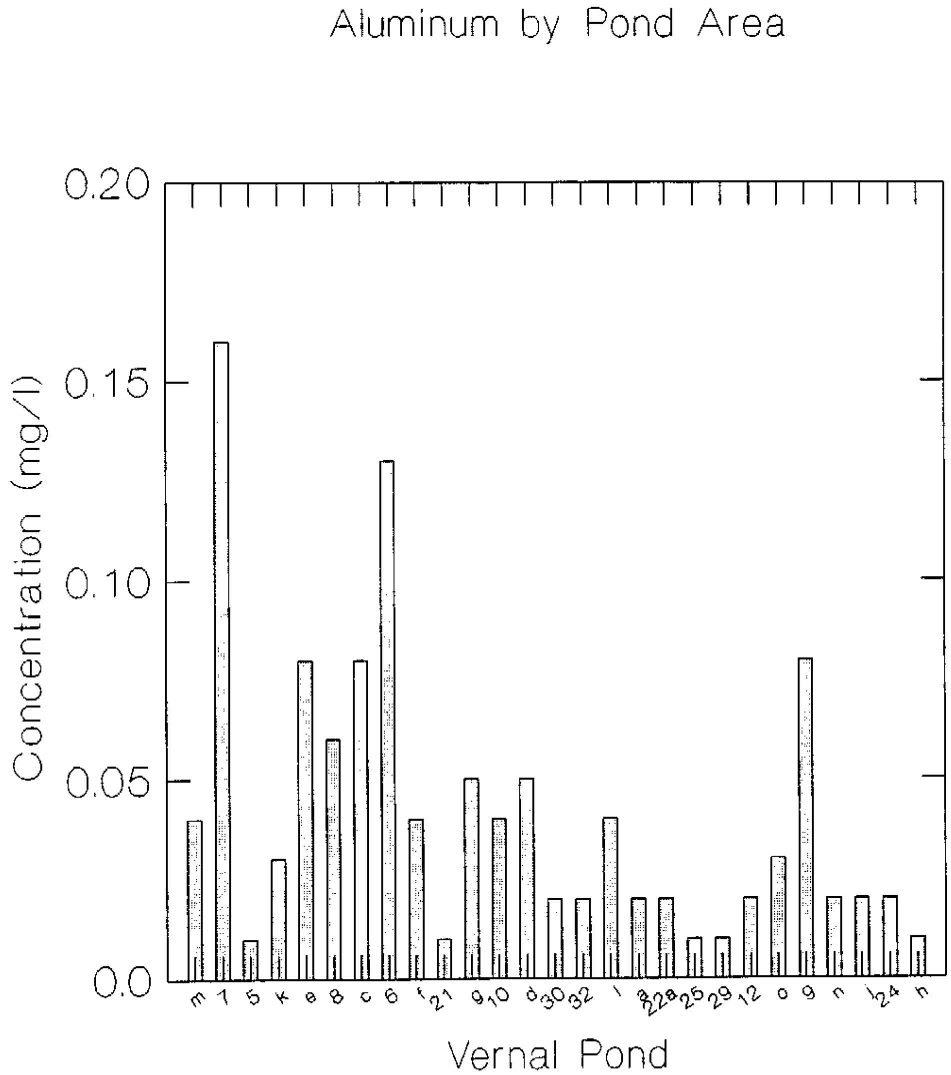
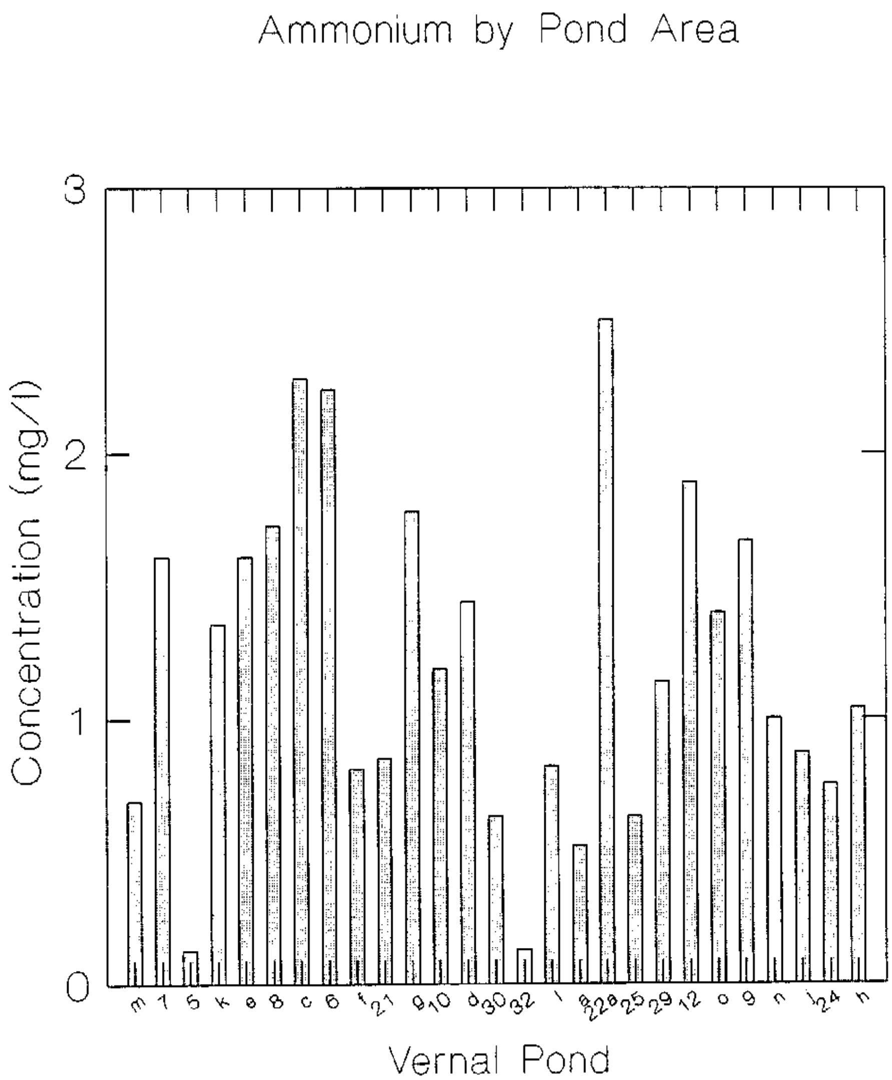

Aluminum by Pond Area
0.20
0.15
0.10
0.05
0.0
Concentration (mg/l)
Vernal Pond
m
7
5
k
e
8
c
6
f
21
g
10
d
30
32
l
a
22a
25
29
12
o
9
n
j
24
h

Figure 6



Ammonium by Pond Area
Concentration (mg/l)
0
1
2
3
m
7
5
k
e
8
c
6
f
21
g
10
d
30
32
l
a
22a
25
29
12
o
9
n
j
24
h
Vernal Pond

Figure 7

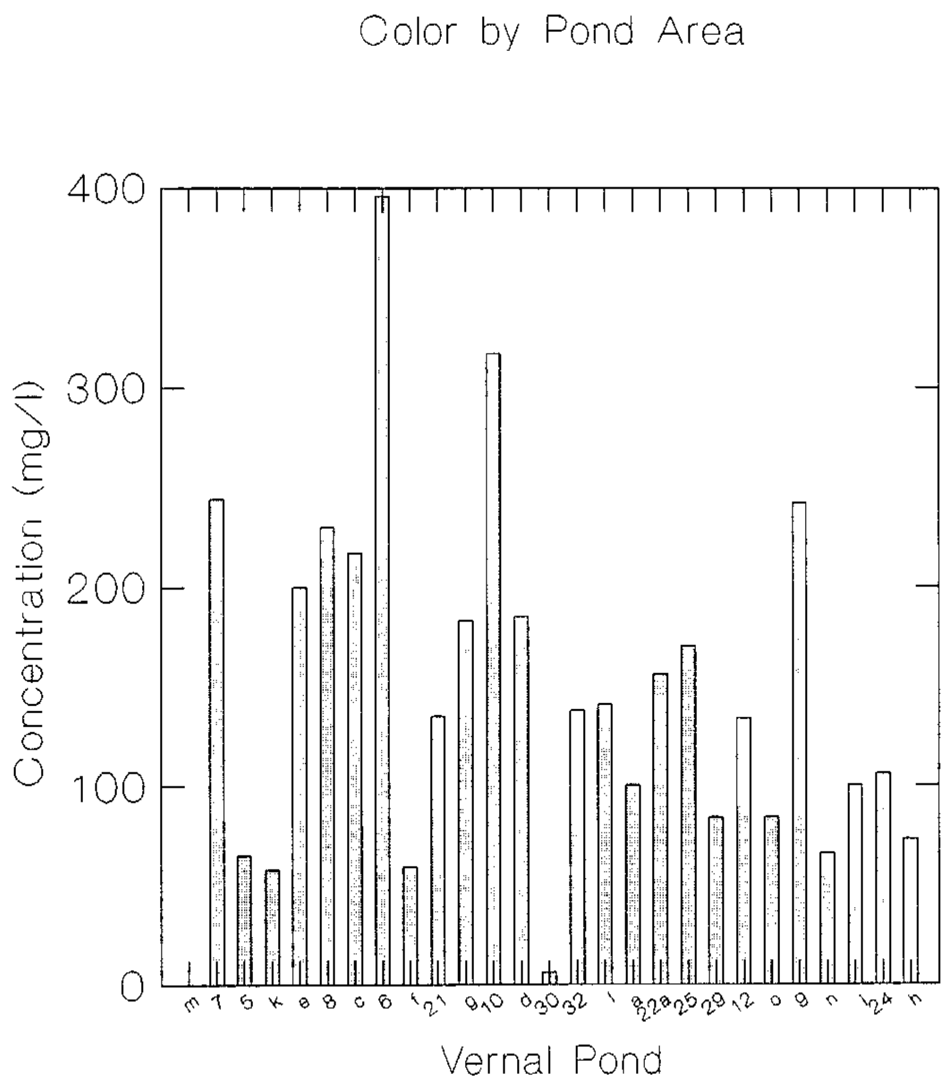

Color by Pond Area
Concentration (mg/l)
400
300
200
100
0
m
7
5
k
e
8
c
6
f
21
g
10
d
30
32
l
a
22a
25
29
12
o
9
n
i
24
h
Vernal Pond

Figure 8

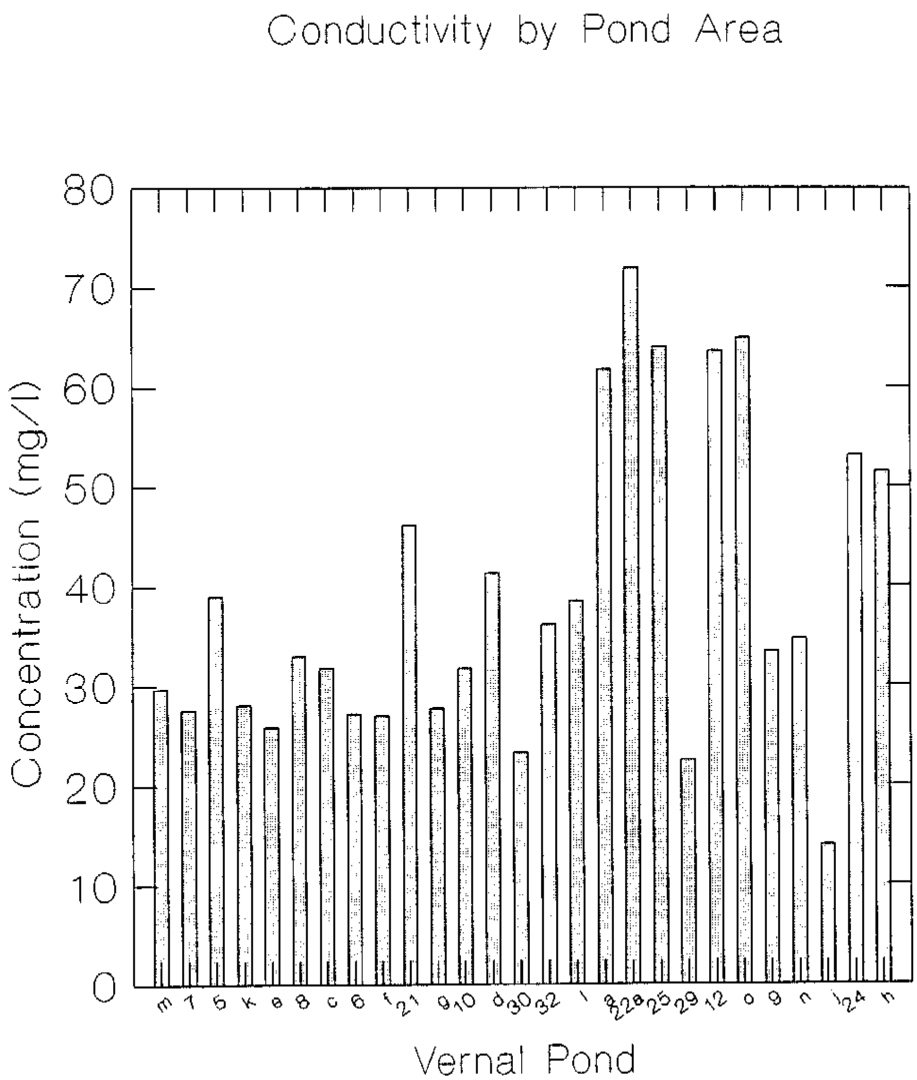
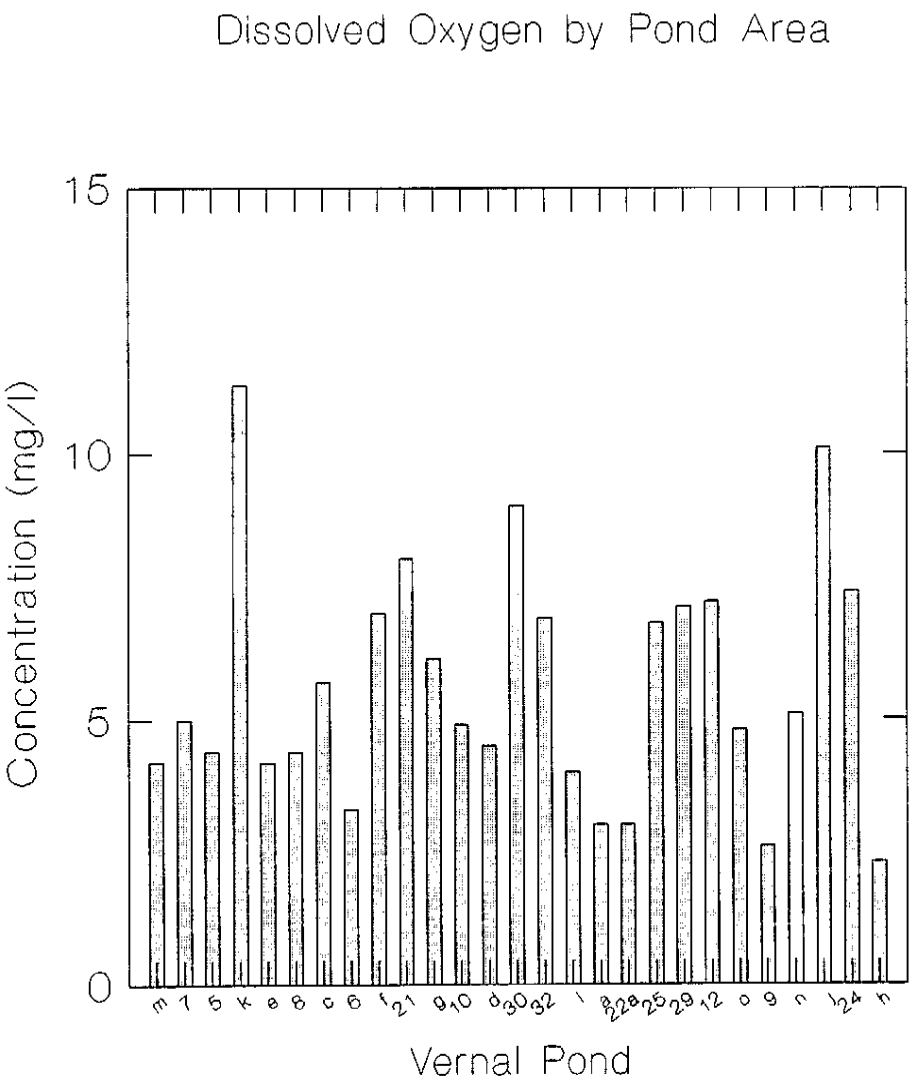

Conductivity by Pond Area
Concentration (mg/l)
0
10
20
30
40
50
60
70
80
m 7 5 k e 8 c 6 f 21 g 10 d 30 32 l a 22a 25 29 12 o 9 n j 24 h
Vernal Pond

Figure 9



Dissolved Oxygen by Pond Area
15
10
5
0
Concentration (mg/l)
m
7
5
k
e
8
c
6
f
21
g
10
d
30
32
i
a
22a
25
29
12
o
9
n
j
24
h
Vernal Pond

Figure 10

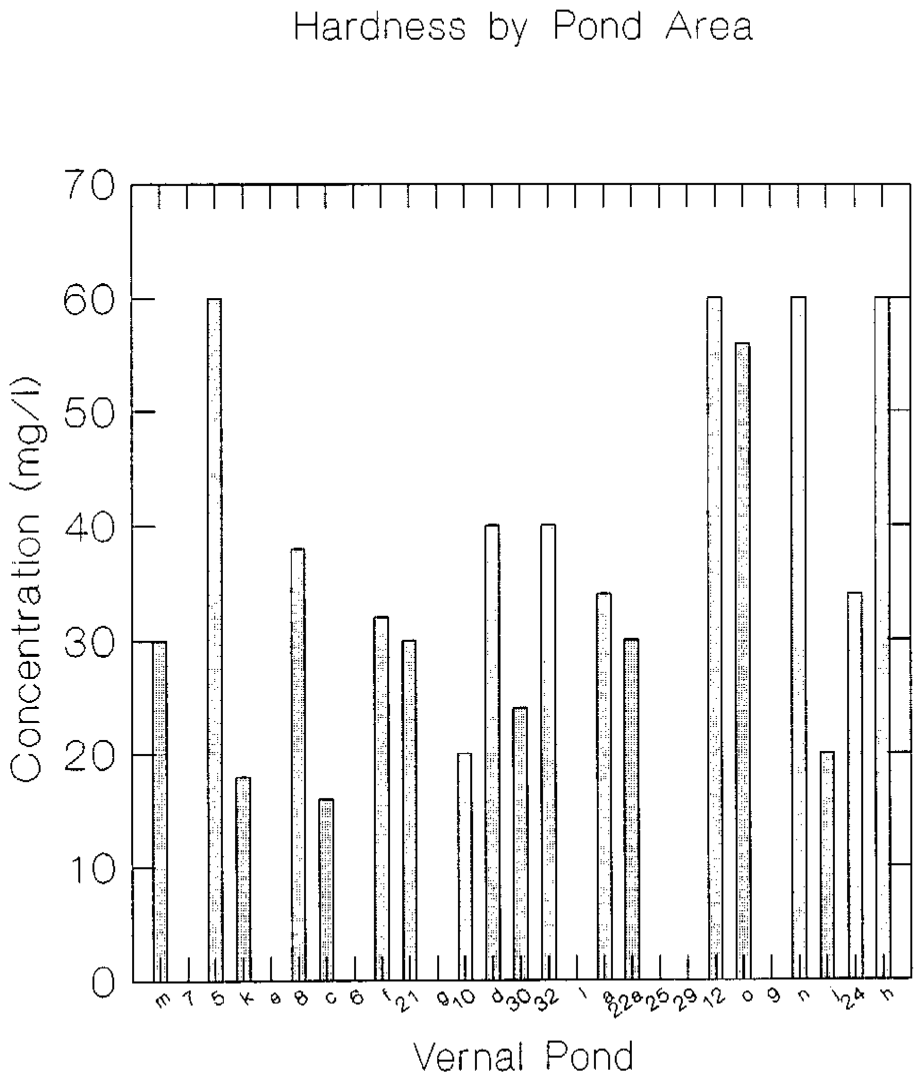

Hardness by Pond Area
70
60
50
40
30
20
10
0
Concentration (mg/l)
m 7 5 k e 8 c 6 f 21 g 10 d 30 32 l a 22a 25 29 12 o 9 n j 24 h
Vernal Pond

Figure 11

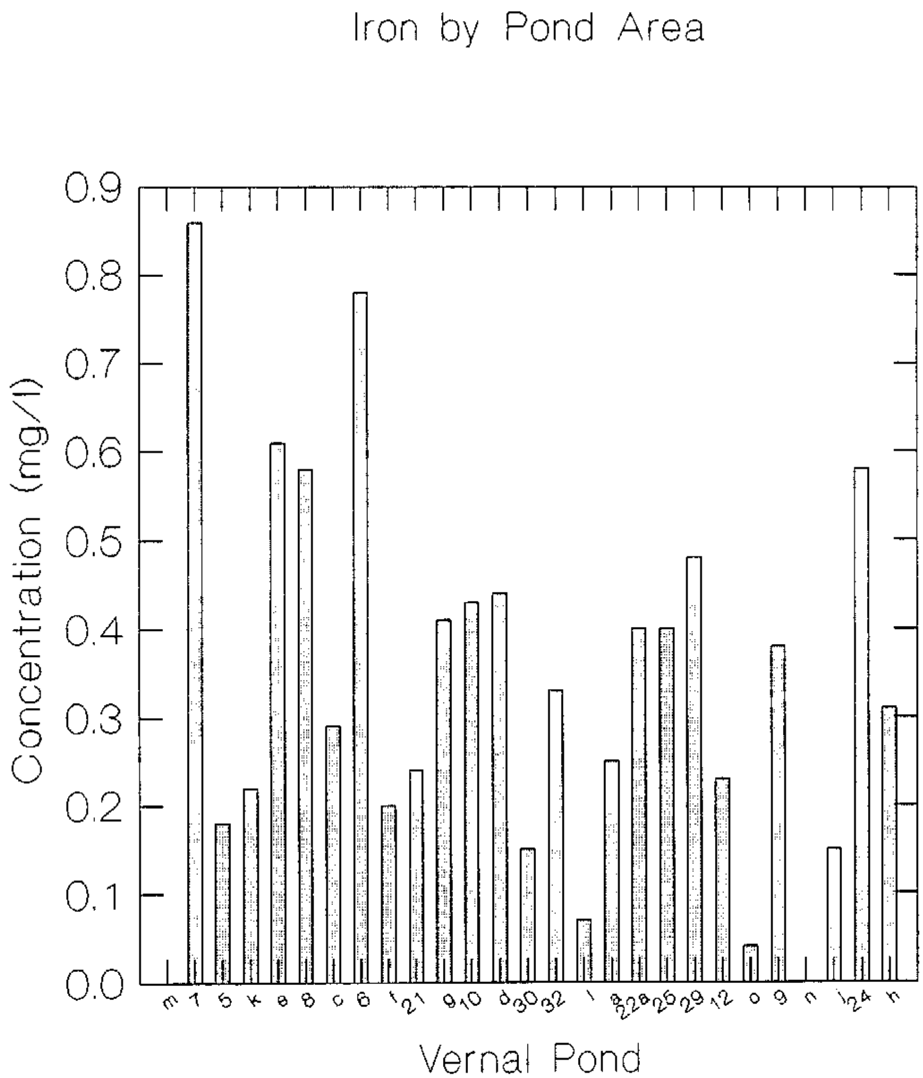

Iron by Pond Area
Concentration (mg/l)
0.9
0.8
0.7
0.6
0.5
0.4
0.3
0.2
0.1
0.0
Vernal Pond

Figure 12

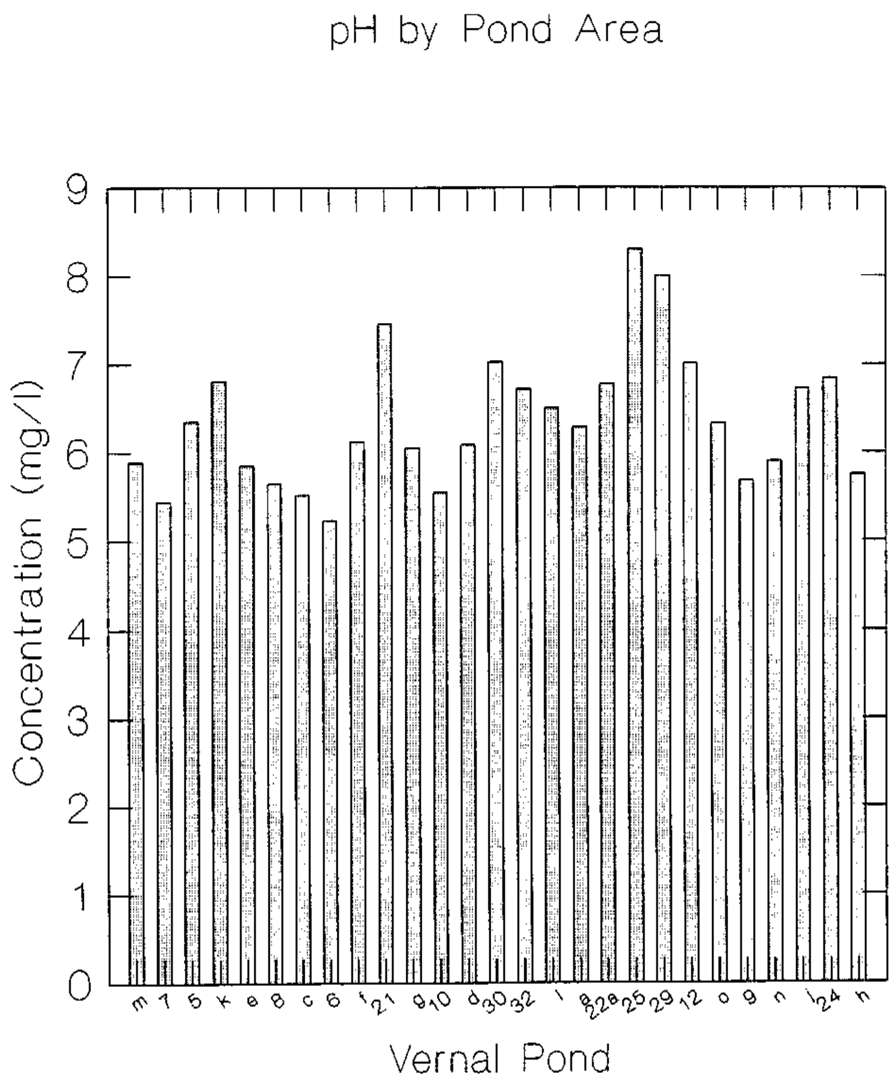

pH by Pond Area
Concentration (mg/l)
0
1
2
3
4
5
6
7
8
9
m 7 5 k e 8 c 6 f 21 g 10 d 30 32 l a 22a 25 29 12 o g n i 24 h
Vernal Pond

Figure 13

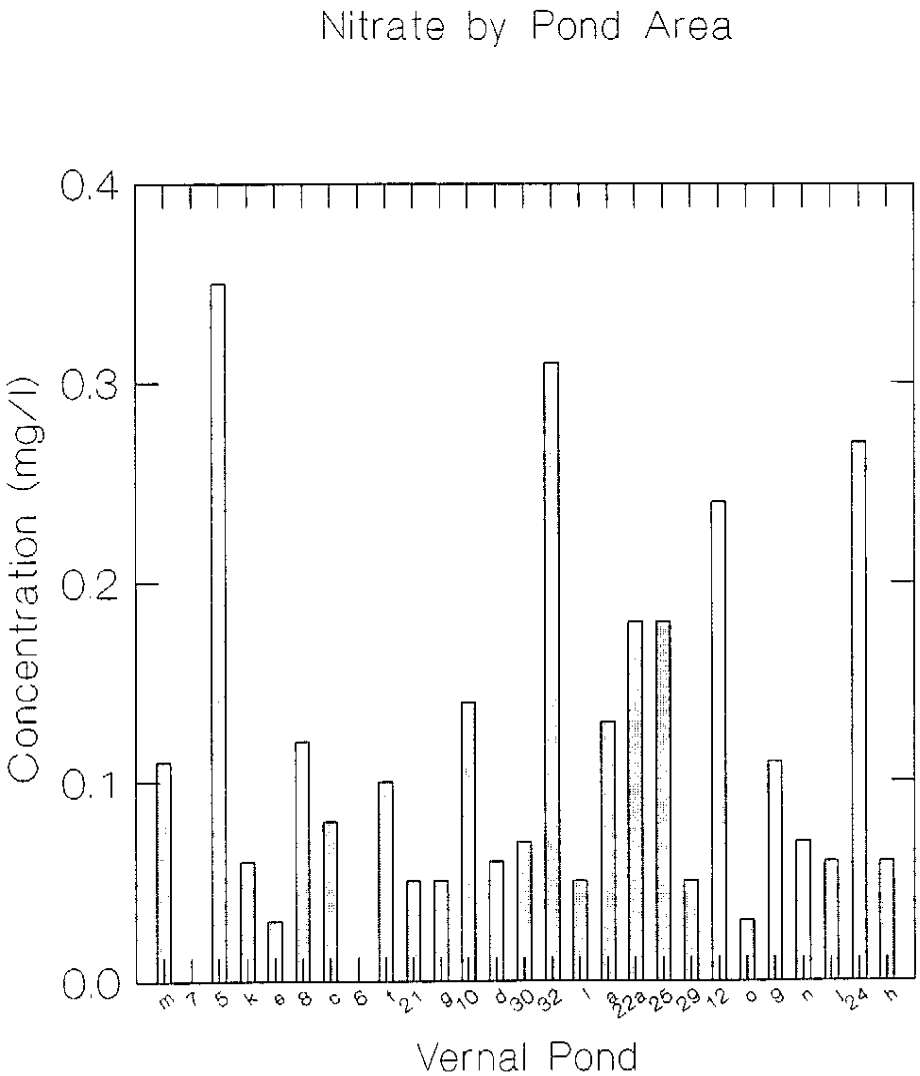

Nitrate by Pond Area
Concentration (mg/l)
0.0
0.1
0.2
0.3
0.4
m
7
5
k
e
8
c
6
21
9
10
d
30
32
22a
25
29
12
o
9
n
24
h
Vernal Pond

Figure 14

% O2 Saturation by Pond Area

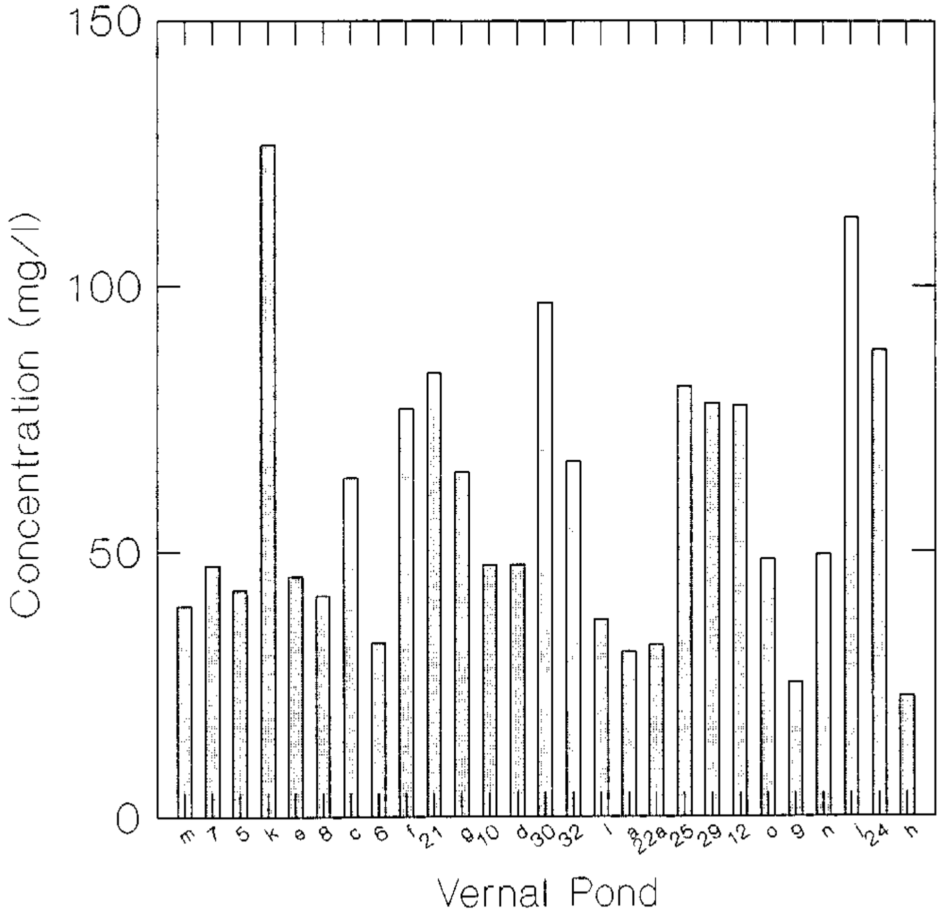


Figure 15

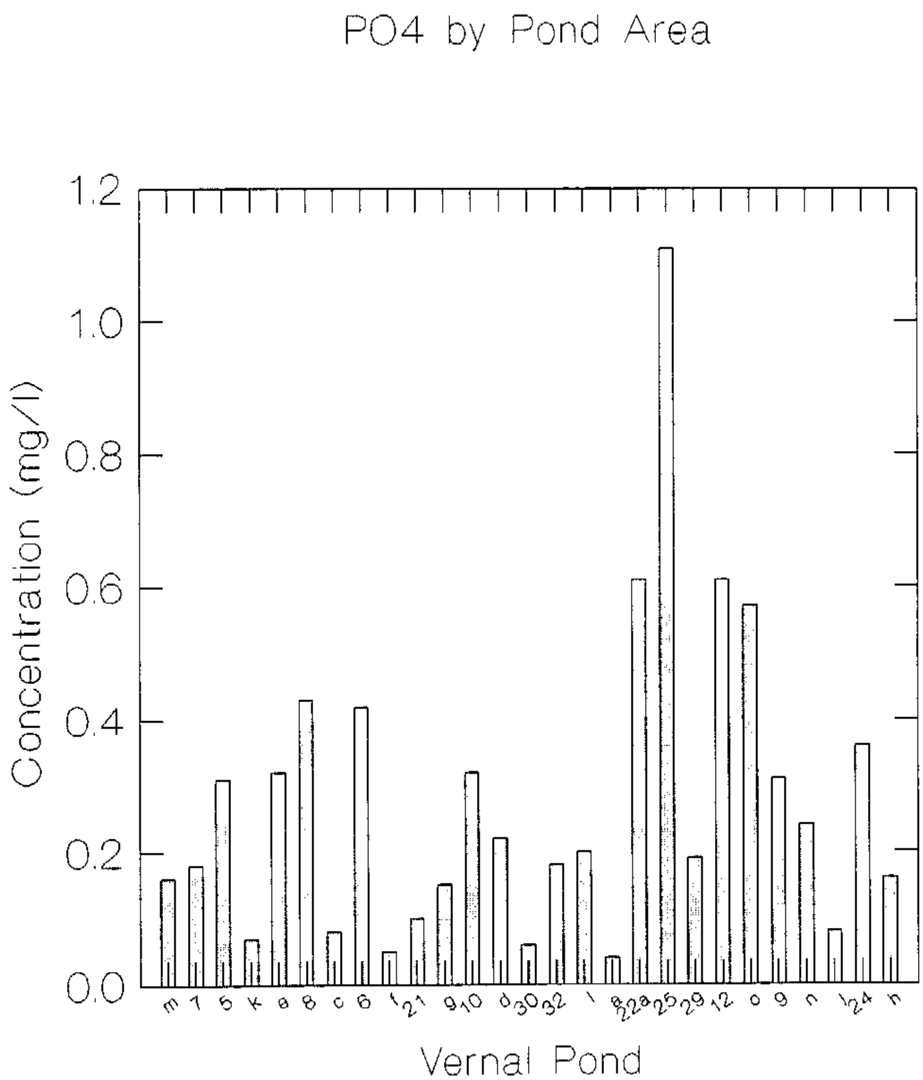

PO4 by Pond Area
Concentration (mg/l)
1.2
1.0
0.8
0.6
0.4
0.2
0.0
m 7 5 k a 8 c 6 f 21 g 10 d 30 32 l e 22a 25 29 12 o 9 n i 24 h
Vernal Pond

Figure 16

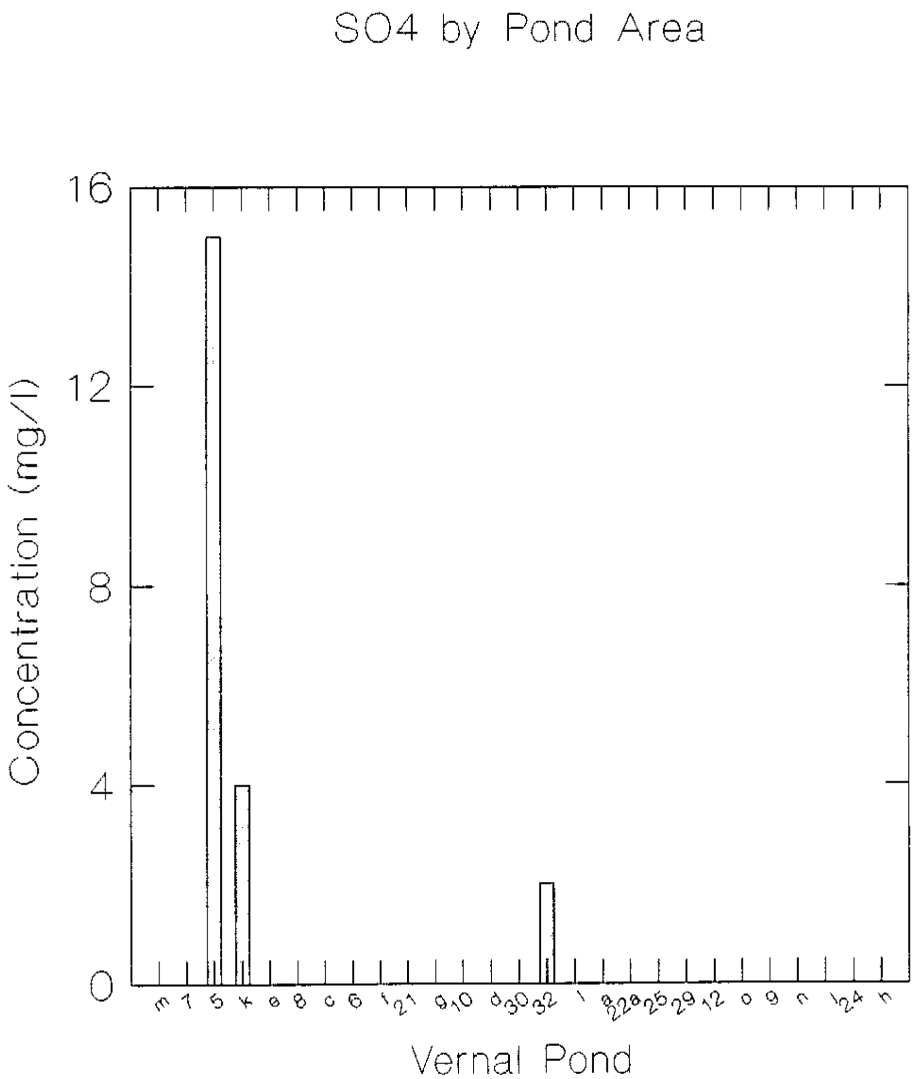
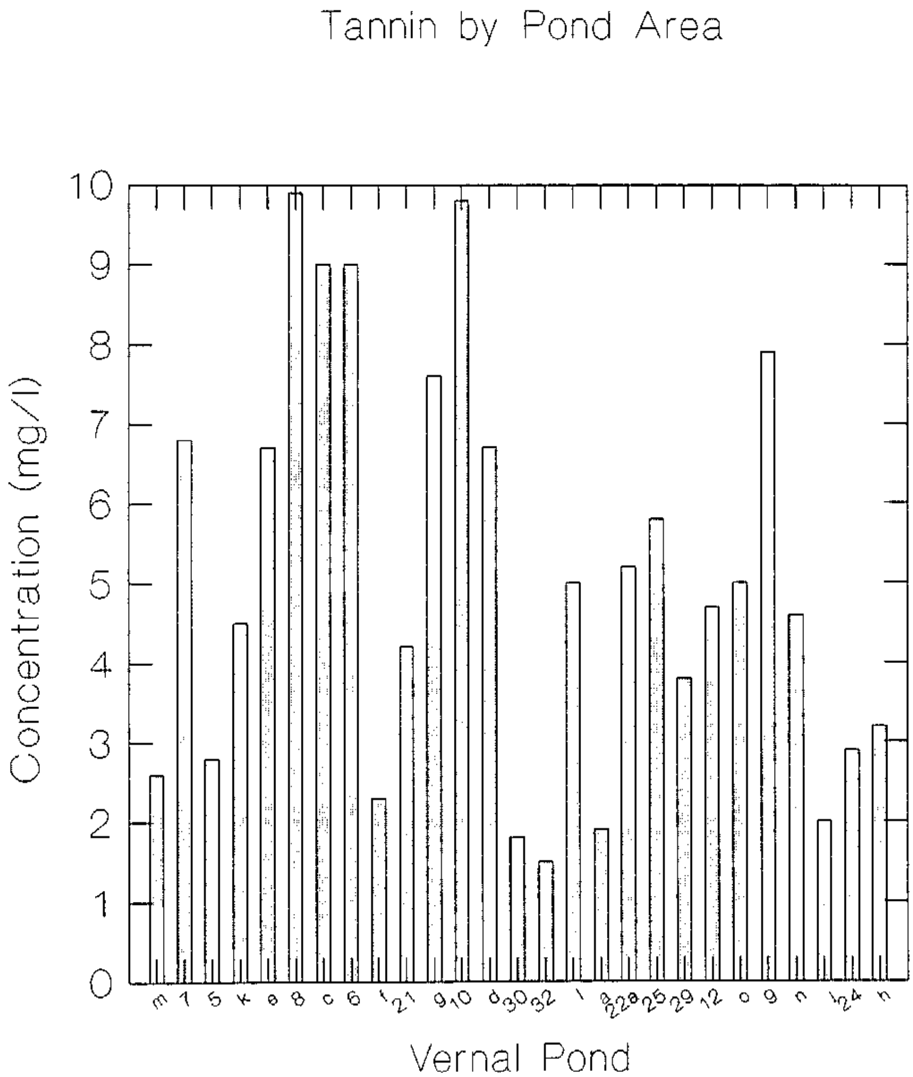

SO4 by Pond Area
Concentration (mg/l)
0
4
8
12
16
m 7 5 k e 8 c 6 f 21 g 10 d 30 32 l a 22a 25 29 12 o 9 n j 24 h
Vernal Pond

Figure 17



Tannin by Pond Area
Concentration (mg/l)
0
1
2
3
4
5
6
7
8
9
10
m
1
5
k
e
8
c
6
f
21
g
10
d
30
32
l
a
22a
25
29
12
o
9
n
i
24
h
Vernal Pond

Figure 18

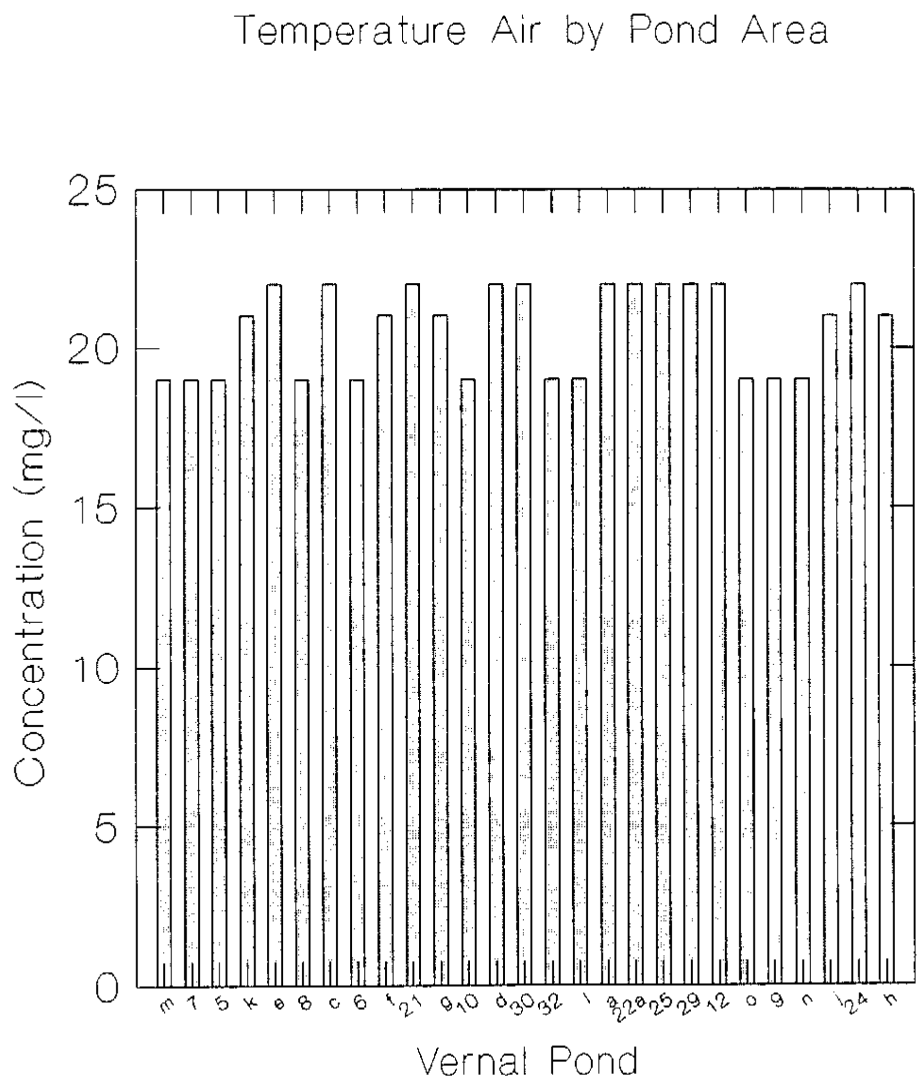

Temperature Air by Pond Area
Concentration (mg/l)
25
20
15
10
5
0
Vernal Pond

Figure 19

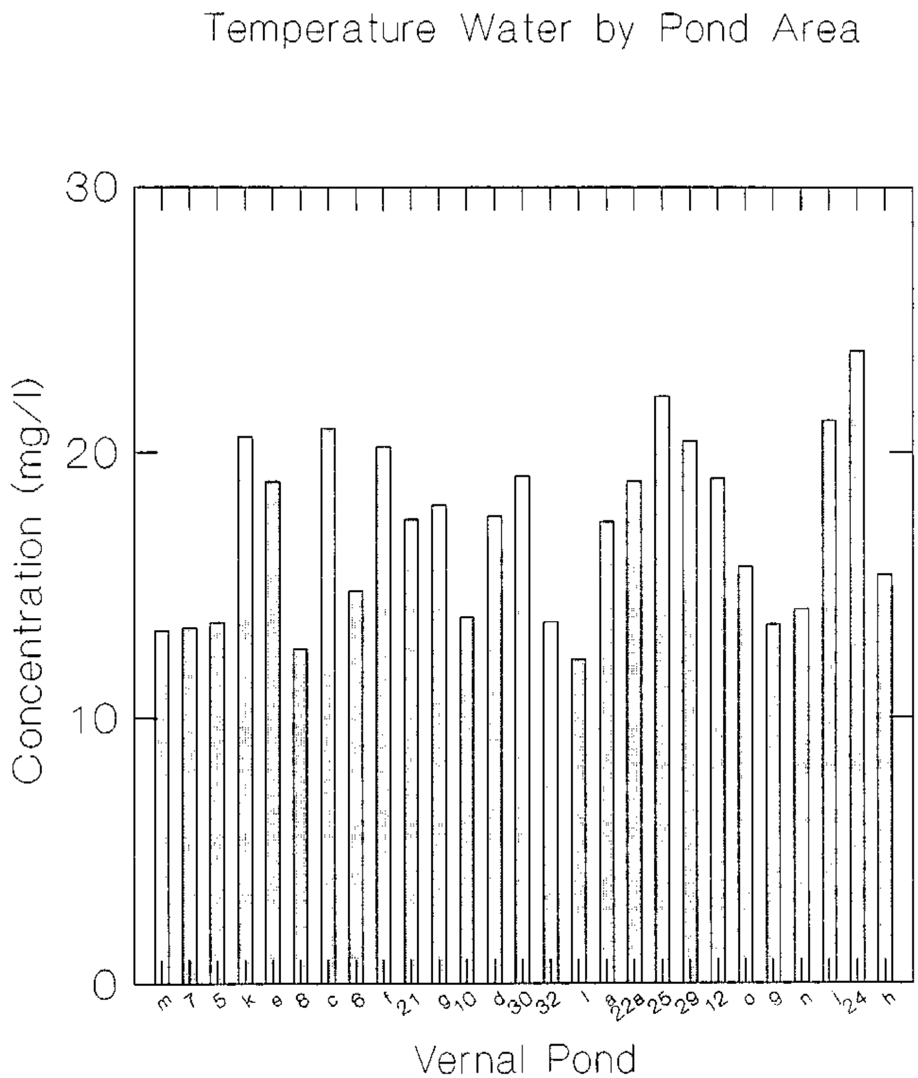

Temperature Water by Pond Area
30
20
10
0
Concentration (mg/l)
m 7 5 k e 8 c 6 f 21 g 10 d 30 32 i a 22a 25 29 12 o 9 n l 24 h
Vernal Pond

Figure 20

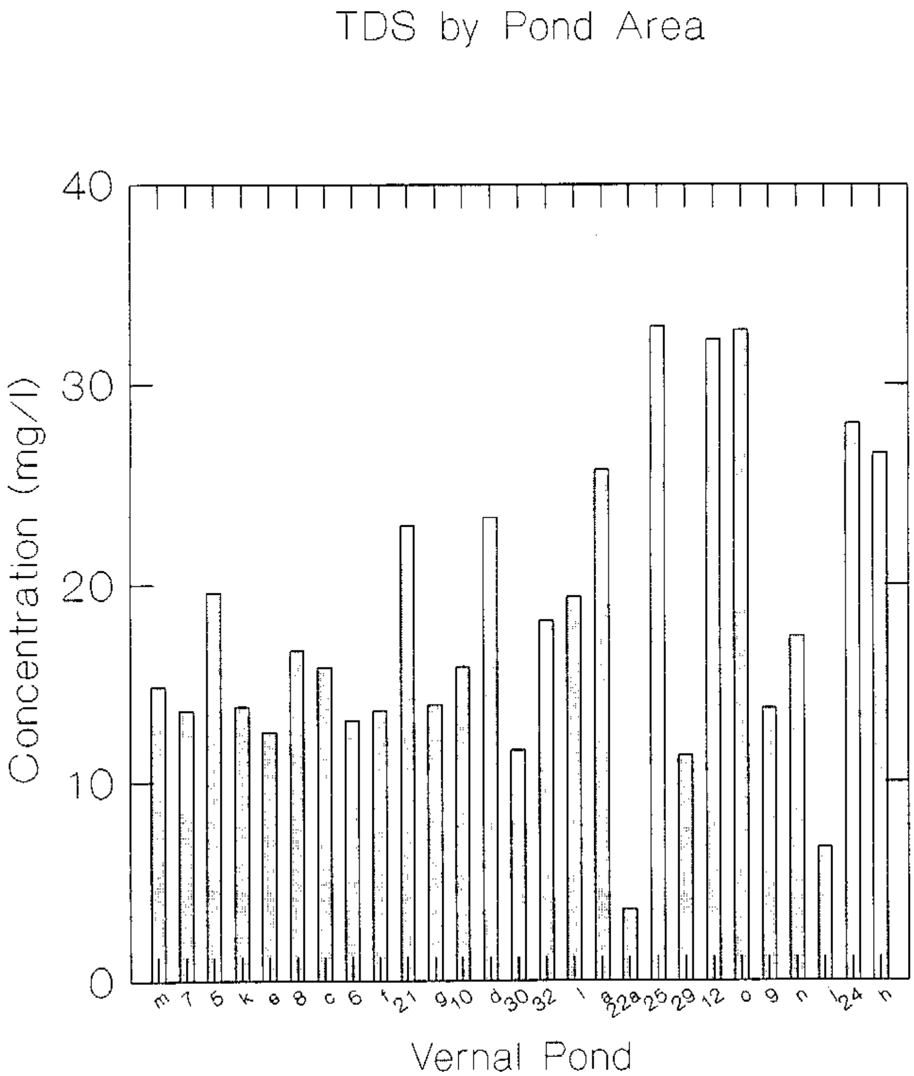

TDS by Pond Area
Concentration (mg/l)
0
10
20
30
40
m 7 5 k e 8 c 6 f 21 g 10 d 30 32 l a 22a 25 29 12 o 9 n i 24 h
Vernal Pond

Figure 21