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Final Report

LAKE MEAD MONITORING PROGRAM

Submitted To

Clark County Sanitation District No. 1
Waste Treatment Physical Development Section

By

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INTRODUCTION

The Lake Mead monitoring program has developed a substantial body of information on the physical, chemical and biological limnology of Lake Mead since 1972. This report summarizes pertinent aspects of that data, with emphasis on studies completed in 1975-76. It is our continuing hope that the data developed by us will be useful to an ever broader group of users of the water resource represented by Lake Mead. We have been privileged to see our data have a significant influence in some very important water resource decisions over the past four years. There is every reason to expect that as the data base builds, its usefulness to user groups will also build.

Past water resource decisions in which our studies have played a significant role were:

1. The decision by the Nevada State Legislature to create the Clark County Wastewater Management Agency, an agency later assimilated into the Clark County Sanitation District.
2. The decision by the Nevada State Legislature to recommend that the Wastewater Management Agency examine the possibilities of developing maximum beneficial in-valley use of water heretofore defined as wastewater.

3. The decision to modify existing water quality standards for Las Vegas Wash to an extent that permitted evaluation of the construction of an advanced wastewater treatment plant as a viable alternative means of handling waste waters from Las Vegas.
4. The definition of a point in Las Vegas bay at which waters from Las Vegas Wash would usually be mixed with waters in Las Vegas Bay. This point is currently defined as the point at which samples can be taken to determine compliance with water quality criteria developed for the Lower Colorado River.

Data and interpretations developed from the data have also been used in a number of other ways; perhaps most notable of these was the documentation necessary to show that conditions in Lake Mead, contrary to the prevailing public opinion, could not have been responsible for the deaths of several dogs along its shores. It was subsequently shown that the dogs died of poisoning at the hands of some unidentified person. Less spectacular but perhaps of greater importance in the long run have been the requests received from other user agencies to apply our familiarity with the limnological conditions in Lake Mead to specific management needs. To date we have undertaken this

kind of specific study, based on our more general data base, for the National Park Service, and the Nevada Department of Fish and Game and the Environmental Protection Agency. In addition, information has been supplied to the Alfred Merritt Smith water treatment plant. The limnological studies, largely supported by Clark County, have thus attracted a broadened base of support as additional needs for related information have been identified.

The primary purposes of the monitoring program have been to develop and maintain a data base essential to the continuing evaluation of the success of the efforts to clear up Las Vegas Wash and to provide a continuing evaluation of the validity of proposed actions. We have developed such a data base and need only to maintain it current for our continuing evaluation to be effective. This data base, however, is also useful and should increasingly be used for other management purposes as well. We are in a position to assist in the rational definition and description of problems related to water resource management in Lake Mead.

SUMMARY

Lake Mead is a deep, subtropical, monomictic lake with surface water temperatures ranging from 10.5° C to 27° C. Thermal stratification develops in May and June, and a classical thermocline becomes established between 10 and 15

meters in July. A turnover begins in October and the lake is completely destratified in January and February. Turnover is weak due to surface temperature only approaching hypolimnetic temperatures.

A negative hetrograde oxygen profile develops with thermal stratification and is found in all reaches of Lake Mead. The most severe oxygen depletion occurs in Las Vegas Bay as a result of higher eutrophic conditions. Oxygen depletion is always associated with the thermocline and hypolimnetic oxygen levels remain high with only minimal oxygen loss during summer stratification.

Our data shows that metalimnetic oxygen depletion is the result of biological respiration. This is supported by the vertical distribution of pH which also has minimum values in the metalimnion, indicating higher concentrations of carbon dioxide being produced in this zone. Phytoplankton and zooplankton respiration are the primary causative agents of the metalimnetic oxygen depletion, accounting for 57 to 94% of the oxygen lost. The primary factor permitting the negative hetrograde oxygen profile is the depth of the lake. Only the upper portion of the hypolimnion is affected by metalimnetic oxygen depletion. Oxygen concentrations in the deeper waters remain high and result in the negative hetrograde oxygen profile.

Phosphorus concentrations throughout Las Vegas Bay, because of nutrient enrichment from Las Vegas Wash, were higher than those found in the Boulder Basin. Phosphorus loading from Las Vegas Wash has increased since 1972 from a mean daily rate of 524 Kg/day (1155 lb/day) to 792 kg/day (1746 lb/day) in 1975. The phosphorus loading from Las Vegas Wash is extremely high when compared with the Colorado River, which, with 200 times the volume, discharges only 950 (2094 lb/day) into Lake Mead. Phosphorus loading for Boulder Basin is in the proposed "dangerous" level. A reduction in the phosphorus loading from Las Vegas Wash to 360 kg/day (794 lb/day) would result in a "permissible" loading rate in Boulder Basin.

Nitrogen appears to be limiting, at least to the numerically dominant dicyclic diatom Cyclotella, during mid-summer. This is indicated by the dominance of the blue-green alga Anabaena, a possible nitrogen fixer, between the early summer and autumnal pulse of Cyclotella. Nitrate-N is almost completely depleted from the epilimnion at this time. Cyclotella does remain dominant in the estuary of Las Vegas Bay where nitrogen is available via Las Vegas Wash. Fall overturn circulates nitrogen from the hypolimnion into the epilimnion and may be the most important factor behind the autumnal pulse of Cyclotella. Nitrogen to phosphorus (N/P) ratios also show an excess of phosphorus during summer stratification, indicating the possibility of nitrogen limitation.

Although phosphorus loading has increased, phytoplankton numbers and biomass have decreased since 1972. This may be related to lake elevations. In 1972, the water level was about 6 meters lower than it was in 1974 and 1975. The higher water levels increased the volume of the inner Las Vegas Bay alone, by approximately $50 \times 10^6 \text{m}^3$. The increased volume would result in a greater dilution of the Las Vegas Wash influent, thereby decreasing the availability of the nutrients to the phytoplankton.

If nitrogen is limiting to most of the phytoplankton, phosphorus loading will have to be reduced to a point where phosphorus becomes limiting before a decline in phytoplankton populations occurs. The nuisance blue-green algae Anabaena should be directly affected by a reduction in phosphorus. If Anabaena is a nitrogen fixer, phosphorus would appear to be the most likely controlling factor, therefore, a reduction in phosphorus should result in lower numbers of Anabaena.

Phytoplankton counts were typically dominated by early summer (June and July) and autumnal (October-December) pulses of diatoms. The blue-green algae Anabaena is usually dominant in September between the diatom pulses. Numbers are always higher in the inner bay because of nutrient enrichment from Las Vegas Wash. The diatom Cyclotella is responsible for both the early summer and autumnal pulses. The early summer pulse appears to originate in the inner bay and expands outward in a wave-like manner on succeeding days throughout the middle and outer bay. The autumnal pulse always occurs throughout Las Vegas Bay and

Boulder Basin in October and is associated with mixing. The early summer pulse of Cyclotella has declined since 1972 and has been accompanied by an increase in the Anabaena population. In 1975, there was no apparent early summer pulse in the outer bay and Boulder Basin apparently because of the complete dominance of Anabaena. This may have resulted from the 4 July flash flood which damaged the Las Vegas City Sanitation Plant and discharged a large quantity of nutrient rich sediments from the Las Vegas Wash marsh system into Las Vegas Bay. However, the dominance of Anabaena may be related to other physical or biological factors.

The early summer and autumnal pulses of Cyclotella have relatively little influence on biomass determinations. Cyclotella never comprised more than 15% of the total biomass of the phytoplankton based on cell volumes. Anabaena was more important, accounting for 15 - 50%. Fragilaria, Glenodinium, Ceratium, and Oocystis were the dominant organisms on the basis of cell volumes. Cyclotella and other diatoms occurring during the autumnal pulse may be important in the taste and odor problems experienced by the Alfred Merritt Smith water treatment plant in the fall.

Primary productivity estimations were extremely high, especially in the inner bay. Lake Mead has been classified as a polluted, eutrophic lake based on primary productivity indices. These indices are based on temperate lakes and do not take into account the extended growing season in a subtropical lake such as Lake Mead. Primary productivity in Boulder Basin is comparable to other tropical or sub-tropical

lakes and conditions in Boulder Basin are probably not as serious as has been reported. The extremely high primary productivity in Las Vegas Bay is of concern as it is indicative of the enriched conditions which have developed because of Las Vegas Wash. Based on maximum phytoplankton volumes, Las Vegas Bay is highly eutrophic and Boulder Basin is mesotrophic.

The limnetic zooplankton community of Boulder Basin generally shows characteristics similar to typical limnetic communities reported in the literature. Rotifers, cladocerans, and copepods are the main components of the zooplankton community. The total population shows three distinct peak periods of abundance, October/November, January/February, and June/July.

The vertical distribution of the summer zooplankton community is quite different from that seen in winter populations. The summer populations exhibited a preference for the upper layers of the water column. The abundant grazers (copepod nauplii and juvenile instars, and cladocerans) showed a definite affinity for the metalimnion (10-25 meters). The presence of a large metalimnetic area appears to set up an energy-subsidy system. The density layering caused by the thermal gradient may allow these organisms to remain in this region, expending little energy because of the constant rain of phytoplankton and detritus from the epilimnion.

Coliform populations in Las Vegas Bay were generally low, however, after the 4 July flash flood higher concentrations were found. Most of the enteric bacteria at that time were found to be Erwinea herbicola and Klebsiella pneumoniae. E. herbicola is generally associated with plant galls and K. pneumoniae is found in association with root systems as well as with fecal material. Because such a large volume of water came down the wash during the flash flood, and because the largest number of enteric organisms found may have been associated with plants, the source of these bacteria may have been from the marsh system and not from the sanitation plants. Salmonella was also detected in Las Vegas Bay after the flash flood.

A laboratory study was conducted to determine the survival of sediment-bound coliforms. Results showed that coliforms do survive for significant periods of time in the sediments and that coliform populations will increase in size in nutrient rich sediments. Since a large fraction of the entering bacterial load is deposited in the nutrient rich sediments at the mouth of the wash, re-suspended bottom sediments may be a significant source of coliforms in Las Vegas Bay.

Studies of current distribution of water from Las Vegas Wash into Las Vegas Bay showed that a density current exists just above the mud-water interface for some distance into the lake. Both bacterial tracers and fluorescent dye tracer in water from Las Vegas Wash were detectable in this density current.

Arrival time of the dye peak was coincident with the bacterial tracer arrival. The transit time from dye injection at North Shore Road to Sample Point 1, 1200 meters downstream, was 45 minutes. Transit time from Sampling Point A to Sampling Point G, 400 meters downstream, was 110 minutes. From G to Sampling Point 2, a distance of 1000 meters, was 245 minutes.

Oxidase negative bacteria in significant numbers over those already present in the lake have been detected as far into the lake as Sampling Point 3 under special conditions (i.e., during a period when the sanitation plant was inoperative) but at Sampling Point 2 on most occasions. In general, the distribution of oxidase negative bacteria correlates well with the arrival of injected dye peaks at sampling points in Las Vegas Bay.

Difficulty in locating relatively low quantities of dye favors the use of indigenous bacteria as current tracers. This is so because the indigenous bacteria in Las Vegas Wash are injected over a rather long period of time in relatively high concentrations. The "component ratio" concept of bacterial tracing has great promise as a means of tracing water distribution patterns of streams into lakes. The utility of this new concept lies in that it is not dependent on the sporadic occurrence of a unique bacterium that is indigenous to the influent stream and not the lake. This new concept, rather,

is dependent on the fact that the relative ratios of bacterial genera in an inflowing stream are maintained as the current from that stream penetrates progressively into the lake. The maintenance of the component ratio makes bacteria occurring in the distinctive current independent of the "noise" created by populations of like bacteria in the lake. Simply, this means that current tracing may be initiated any time the need arises.

For the oxidase negative bacteria, standard methods result in misleading interpretations because the toxicity of inhibitors present in the media results in heavy mortality during incubation of samples. This has proven to be the key barrier to the use of oxidase negative bacteria as tracers of water distribution patterns.

For the oxidase negative bacteria, the multitest scheme for identification provides a source of descriptors, once the bacteria have been isolated. The entire array of descriptors is too cumbersome for routine use, as is the sequential treatment of individual single isolates. A single solution to the inhibitory initial isolation and the isolate identification problems has been resolved. This solution involves initial plating on non-inhibitory agar followed by serial replica plating onto multi-descriptor media, a measure which allows simultaneous evaluation of similar characteristics and percentage distributions of hundreds of bacteria at once rather than one at a time.

Location of Sampling Stations

In 1972, 14 stations were located throughout Las Vegas Bay. The mid-channel stations were retained in 1974, and 3 additional stations (Stations 6-8) were located in the Boulder Basin (Fig. 1). In 1975, regular sampling at stations 7 and 8 was discontinued. Additional samples were taken at other locations in the Boulder Basin and in the upper basins at various times.

Samples were taken weekly at each of the 14 regular stations in 1972. In 1974 and 1975, samples were taken monthly during winter and spring or bi-weekly during the summer and fall (June to October).

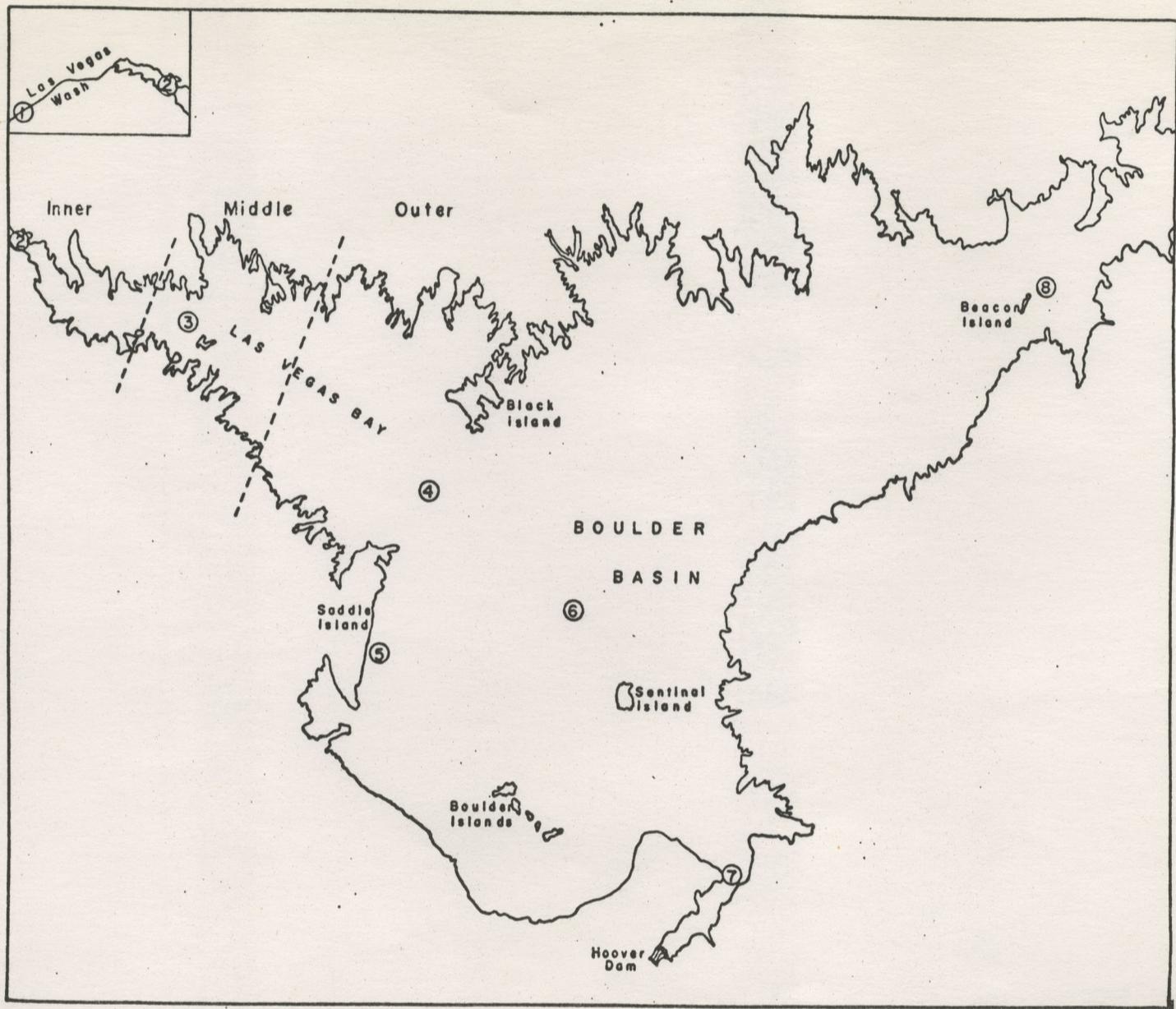


Figure 1. Location of sampling stations in the Boulder Basin

Physical and Chemical Characteristics of Lake Mead

by

John. Baker

Introduction

Regular measurements of temperature, dissolved oxygen, conductivity, pH, and nutrients were taken in 1972, 1974, and 1975 as part of the Lake Mead Monitoring Program. Sampling was mainly confined to the Boulder Basin. Some additional data were obtained from other areas of the lake.

Description of the Study Area

Lake Mead was formed by impounding the Colorado River by Hoover Dam in 1935. The lake is located in the Mohave Desert where maximum temperatures over 40° occur regularly in June through September and winds over 30km/hr are frequent. At the present time, the lake has a maximum depth of over 130 meters. The capacity of the lake, at an elevation of 374 meters above sea level, is $36 \times 10^9 \text{ m}^3$ with a surface area of 660 km^2 (Lara and Sanders, 1970). The average sediment accumulation between 1935 and 1964 was $1.13 \times 10^8 \text{ m}^3/\text{yr}$. This has been substantially reduced by the closure of Glen Canyon Dam above Lake Mead (Hoffman and Jones, 1973). Lake Mead consists of four major basins separated by deep narrow canyons. The major reaches in downstream order are Pierce Basin, Iceberg Canyon, Gregg Basin, Virgin Basin, Boulder Canyon, and Boulder Basin. The Moapa and Virgin Rivers, discharging into the Overton Arm of Virgin Basin,

and Las Vegas Wash, discharging into Las Vegas Bay, a large arm of Boulder Basin, are the only other tributaries to Lake Mead. Investigation of the effects of the Moapa and Virgin Rivers has not been undertaken. Las Vegas Wash is a nutrient rich stream discharging industrial and sewage effluent into Las Vegas Bay.

Materials and Methods

Temperature, dissolved oxygen, conductivity, and pH were measured with a Model IIA Water Quality Analyzer (Hydrolab Corporation) in five meter intervals to a maximum depth of 90 meters. Measurements were taken in one meter intervals through the metalimnion during summer stratification in 1974 and 1975.

Water samples for nutrient analysis were taken at each station at various depths with a three liter Van Dorn water sampler throughout 1974 and 1975. The samples were preserved with mercuric chloride and analyzed for total phosphorus, dissolved phosphorus, ammonia nitrogen, nitrite-nitrate nitrogen, and Kjeldahl nitrogen. Nutrient analyses were performed by the Land and Water Monitoring Branch of EPA, Las Vegas, Nevada.

Results

Temperature - Water Temperatures ranged from 10.5°C in January and February to 27°C in July and August (Table 1).

Table 1. Mean surface water temperatures at Station 4.

	1972	1974	1975
Jan.	10.5		
Feb.	10.5		
Mar.	15.3	15.0	
Apr.	16.0	15.5	13.5
May	19.3	21.5	19.5
June	23.6	24.2	22.0
July	25.3	26.5	27.0
Aug.	25.8	25.7	26.0
Sept.	23.7	24.5	26.0
Oct.	20.8	21.3	20.5
Nov.	17.5	16.8	16.2
Dec.		13.0	14.5

Thermal stratification developed in May and June with the greatest thermal gradient (ca. 3°C change/5 meters) between 5 and 10 meters. In July, a classical thermocline (Hutchinson 1957) became established between 10 and 15 meters when surface water temperatures reached 26°C . The thermocline remained at 10 meters through September in 1974 and 1975, although surface water temperatures were slightly cooler. Mixing usually begins in October but this occurred early in 1972 with mixing to a depth of 15 meters in September. The lake was completely destratified in January and February when surface water temperatures reached 10.5°C . Hypolimnetic water temperatures (90 meters) usually remained constant at 10.5°C and, therefore, turnover was very weak.

Oxygen - With the development of thermal stratification there was a loss of metalimnetic oxygen (Table 2) resulting in a negative hetrograde oxygen profile (Hutchinson, 1957). The loss of metalimnetic oxygen was continuous throughout the period of summer stratification and, therefore, the lowest metalimnetic oxygen levels usually occurred in September just prior to mixing (Fig. 2). In 1974 and 1975, the lake remained stratified at 10 meters through September resulting in a pronounced narrow zone of low oxygen. This narrow zone did not develop in 1972 due to lower water temperatures and mixing which began in late August and September.

Table 2. Temperature ($^{\circ}\text{C}$) and dissolved oxygen (mg/l) at Station 4
April through October 1975.

Depth	28 April		17 June		28 Aug.		2 Oct.	
	Temp $^{\circ}\text{C}$	O_2						
0	13.5	9.6	22.5	10.5	26.0	9.2	25.5	10.4
5	13.0	9.6	22.5	10.5	26.0	9.2	24.5	9.1
10	13.0	9.3	20.5	9.2	25.0	7.0	24.0	8.4
15	12.5	8.9	17.5	7.8	22.5	4.5	24.0	8.0
20	12.0	8.5	15.5	7.4	20.0	3.7	20.0	2.7
25	11.5	8.4	14.0	7.4	18.0	3.6	18.5	3.1
30	11.0	8.1	13.5	7.6	17.0	4.1	17.5	3.1
35	11.0	8.1	12.5	7.6	15.5	4.2	16.5	3.1
40	11.0	8.0	12.0	7.8	14.0	4.4	14.5	4.2
45	10.5	7.9	12.0	7.6	14.0	5.0	13.5	5.1
50	10.5	7.9	12.0	7.5	13.0	5.2	13.0	5.8
60	10.5	7.8	11.0	7.5	12.0	5.6	12.0	5.8
70	10.5	7.8	10.5	7.4	11.5	6.3	11.5	5.7
80	10.5	7.7	10.5	7.6	11.0	6.5	11.0	5.9
90	10.5	7.7	10.5	7.6	11.0	6.6	11.0	6.1

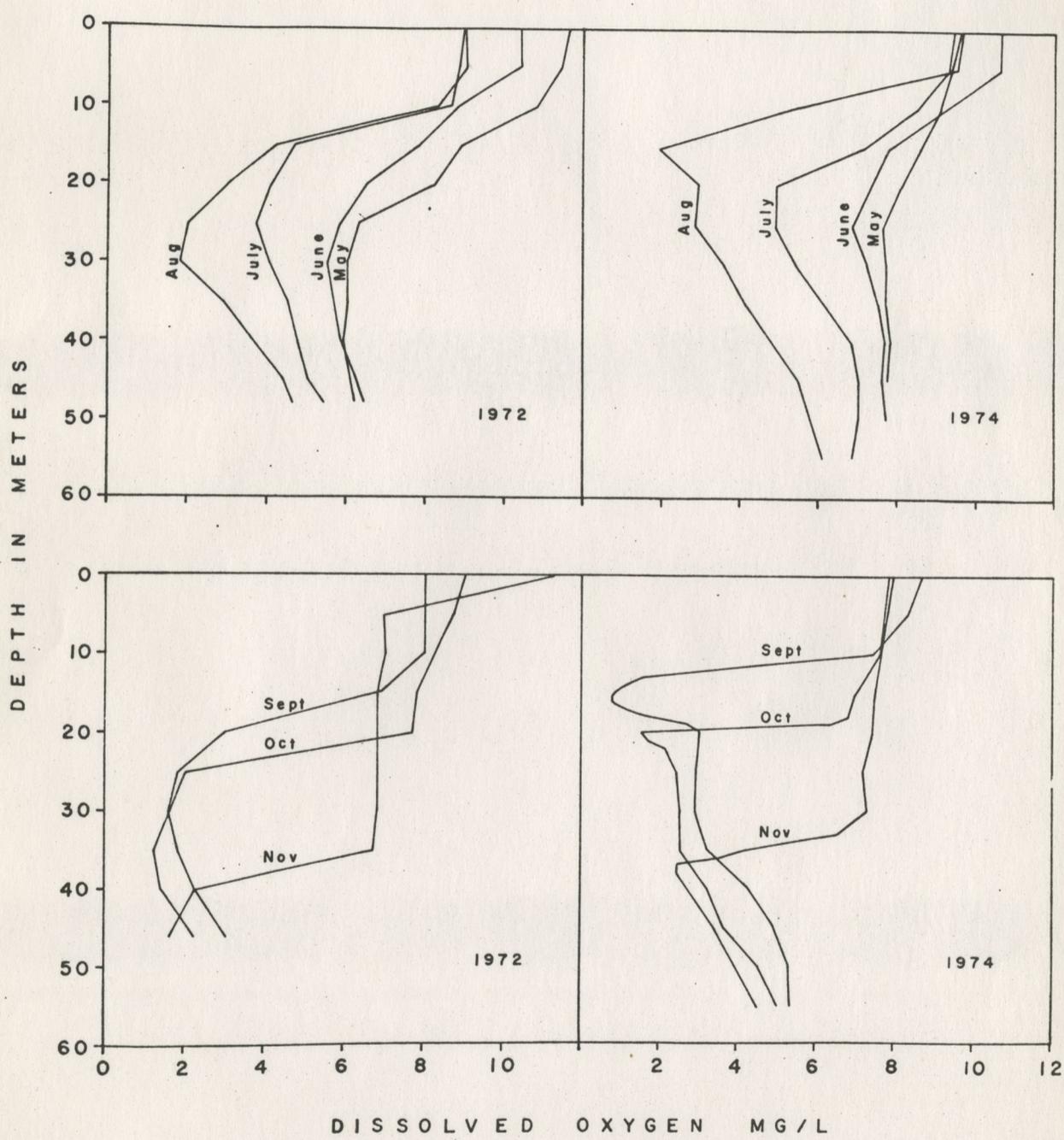


Figure 2. Vertical distribution of dissolved oxygen at Station 5.

The negative hetrograde oxygen profile was found in all reaches of the lake (Table 3), except in the Gregg and Pierce Basins. Las Vegas Bay and Boulder Basin had the most pronounced metalimnetic oxygen depletion. Metalimnetic oxygen levels below 2.0 mg/l in the Boulder Basin and below 1.0 mg/l in Las Vegas Bay were found in August and September. The zone of depletion was broader in Las Vegas Bay than in Boulder Basin. In 1972, the Overton arm of Virgin Basin showed a broad zone of depletion similar to Las Vegas Bay but did not reach comparable low levels.

pH - The hydrogen ion concentration ranged from 7.3 to 8.8. The highest readings were at the surface during the summer and apparently resulted from relatively high phytoplankton production. Vertically, the lowest values were always found within the metalimnion and corresponded with low oxygen levels (Fig. 3). This indicates that respiratory activity is important in both increasing the pH level and reducing the oxygen levels in the metalimnion.

Conductivity - Conductivity was generally constant at 1100 microhoms/cm and did not change vertically except in Las Vegas Bay stations 2 and 3 (Table 4). Higher conductivity levels were found at these stations as a result of a density current or cell from Las Vegas Wash.

Table 3. Vertical distribution of dissolved

Depth	Station 4 23 Aug 72	Black Canyon 23 Aug 72	Virgin Basin 31 Aug 72
0	8.9	8.1	8.5
5	8.9	8.1	8.9
10	7.7	7.9	8.1
15	3.0	6.9	6.8
20	2.7	1.5	5.8
25	1.7	1.3	5.7
30	1.7	1.7	5.6
35	2.5	2.8	5.8
40	3.6	3.6	6.1
45	4.0	4.0	6.4
50	4.3	5.2	6.8
55	5.1	5.8	7.1
60	5.4	5.9	7.0
65	5.4	6.2	7.2
70	5.7	6.3	7.1
75	5.9	6.4	7.1
80	6.0	6.6	7.1
85	6.1	6.6	6.8
90	6.1	6.6	6.7

oxygen at various points in Lake Mead.

Overton Arm Temple Basin Gregg Basin
14 Oct 72 14 Oct 72 14 Oct 72

8.5	9.0	10.4
8.3	8.7	10.2
8.3	8.4	9.8
8.1	8.3	8.7
3.9	6.1	8.6
3.4	6.6	8.3
3.2	6.9	8.1
2.8	4.3	8.0
3.0	4.4	
3.3	5.1	
3.2	5.5	
3.6	5.9	
3.8	6.2	
4.3	6.5	
	6.7	
	6.7	
	6.8	
	6.6	
	6.6	

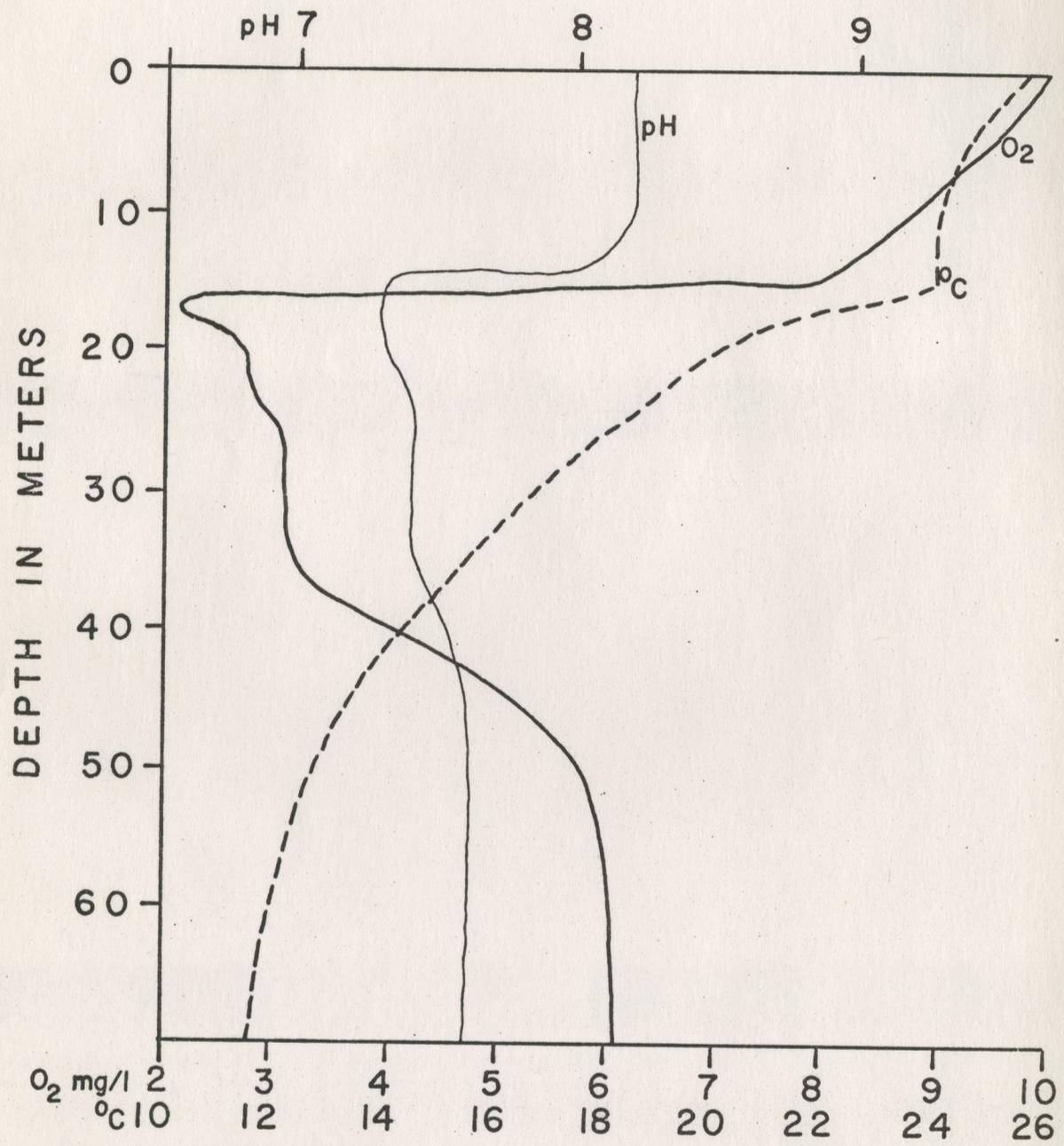


Figure 3. Vertical distribution of temperature, dissolved oxygen and pH at Station 4, 2 October 1975.

Table 4. Conductivity in microhoms/cm at stations 1-3
April 1975 through February 1976.

Date	28 April			17 June			28 Aug.			20 Nov.			22 Dec.			Feb.		
	Station 1	Station 2	Station 3															
0	4200	1100	1100	4300	1150	1100	4500	1200	1100	4050	1100	1100	3800	1100	1100	4200	1050	1050
5		1100	1100		1150	1100		1200	1100		1100	1100		1100	1100		1050	1050
10		2300	1100		2500	1100		1600	1100		1800	1100		1600	1100		2400	1050
15			1100			1300			1600			1100			1100			1050
20			1100			1300			1300			1100			1100			1050
25			1100			1300			1150			1100			1100			1050
30			1100			1200			1200			1100			1100			1050
35			1150			1100			1200			1100			1100			1050
40			1200			1100			1200			1150			1100			1050
45			1600			1100			1200			1400			1400			1600

The density current was located along the bottom when the lake was destratified (November-April). In May, thermal stratification developed and water temperatures increased in Las Vegas Wash, changing the density relationships of the two water masses. At that time, the water from Las Vegas Wash was not as dense as the cooler hypolimnetic water and the density current was located in the area of the thermocline. The density current remained within the metalimnion throughout summer stratification (May - September). There was no evidence, as indicated by higher conductivity levels, of the density current reaching the mouth of Las Vegas Bay (Station 4). There appeared to be substantial mixing and dilution of the current before it reached this point.

Nutrients - Nutrient concentrations for 1975 are presented in Appendix I. Phosphorous and nitrogen were higher and more variable in Las Vegas Bay than in Boulder Basin. This was due to nutrient loading from Las Vegas Wash and was especially evident at Stations 2 and 3 within the density current. A complete discussion of nutrient concentrations will be included in the next section of this report.

Discussion

Lake Mead can be classified as a deep, subtropical, monomictic lake according to Hutchinson (1957). The lake

becomes thermally stratified, although a classical thermocline occurs only for a short period in July. Thermal stratification of a lake is primarily dependent on incident solar radiation but is affected by wind and the degree of exposure of the lake. Therefore, thermal stratification in Lake Mead is not pronounced due to the large surface area of the lake and high summer winds which tend to destratify the lake to some degree.

The occurrence of a negative hetrograde oxygen profile in Lake Mead was first reported by Hoffman et al. (1967). A negative hetrograde oxygen profile was evident in 1944 (unpublished data, Bureau of Reclamation) and low metalimnetic oxygen levels probably have occurred since the formation of the lake. The cause of metalimnetic oxygen depletion in Lake Mead appears to be due to biological respiration. This is supported by the vertical distribution of pH which is a function of carbon dioxide concentrations. In solution, carbon dioxide reacts with water to form carbonic acid, bicarbonate, and carbonate ions. The carbonic acid will lower the pH, although the system is buffered by the bicarbonate and carbonate ions. The lowest pH values were always found in the metalimnion and corresponded with low oxygen levels, thus indicating that biological respiration is occurring on a large scale.

A further discussion on the causes of the metalimnetic oxygen depletion will be made in a separate section of this report.

Effects of Las Vegas Wash, a Nutrient Rich
Stream, on Phytoplankton Populations in
Boulder Basin, Lake Mead, Nevada.

by

John Baker

INTRODUCTION

Phytoplankton succession, biomass and primary productivity were determined to evaluate the effects of nutrient enrichment. Las Vegas Wash is a nutrient rich stream discharging industrial and sewage effluent from the Las Vegas metropolitan area into a narrow estuary at the head of Las Vegas Bay. Las Vegas Wash is the only external enriched nutrient source of any magnitude discharging into Boulder Basin. Therefore, we were able to examine the effects of nutrient enrichment from virtually a single source on a relatively large body of water.

MATERIALS AND METHODS

Phytoplankton - A one liter Van Dorn water sampler was used to collect samples. Each sample was a combination of three separate 1 liter subsamples collected from the same site. Samples were usually taken monthly or biweekly at each of the stations. In 1972, samples were taken weekly. Phytoplankton enumerations were made on samples using a modification of the membrane filter technique (McNabb, 1960). Identifications were made on live material from 200 ml concentrated samples which served as a guide to identifications on the membrane filters. Colonies and filaments were counted as a single unit.

Cell Volumes - Phytoplankton volumes were estimated by determining the average cell volumes for 20 of the most important organisms. The average cell volumes were multiplied by the cell counts to arrive at a total phytoplankton volume.

Chlorophyll - Pigment analyses were made on 500-1000 ml samples filtered through Watman G F/C filters. The filters were pre-treated with $MgCO_3$. The chlorophyll was extracted by grinding the filters in 5 ml of acetone. Pigment concentrations were determined from formulas given by Parsons and Strickland (1963) using data obtained with a Coleman Junior spectrophotometer.

Primary Production - Estimations of primary productivity were based on carbon-14 techniques (Steeman Nielsen, 1951) using modifications of Saunders et al. (1962). Light and dark bottles (pyrex 125 ml) were filled with water collected at the surface, 1, 3, 5, and 10 meters. Each bottle was spiked with 0.5 μCi of $NaH^{14}CO_3$, suspended from a buoy at the depth of collection, and incubated for a period of 4-6 hours. After incubation, the samples were returned to the laboratory in a light-proof box. The samples were filtered through 0.45 μ Millipore filters and the residue on the filters was washed with 10-15 ml of a .005N HCl, 5 percent formalin solution. Metricel filters were used in July - October 1975 but were found to reduce counting efficiency by a factor of 5 times and, subsequently, were not used. The filters were placed in glass

or polyethylene scintillation vials and dried in a dessiccator for 24 hours. A xylene based liquid scintillation solution was added to the vials and the activity of the samples was determined. The activity of the samples was converted to milligrams of carbon per unit area per day using the methods and tables of Saunders et al. (1962).

RESULTS

Phytoplankton Numbers - Phytoplankton counts for the most numerous organisms for April 1975 through March 1976 are presented in Appendix II. Phytoplankton numbers were always higher in the inner bay (Station 2) due to nutrient enrichment from Las Vegas Wash. Dominant organisms during the 3 years of investigation were similar, especially in the summer. Total numbers have decreased since 1972 and the successional patterns have changed.

Winter and early spring (January-April) phytoplankton populations were generally below 300 organisms/ml except at Station 2 where counts over 1000 organisms/ml were found. The phytoplankton populations were usually dominated by diatoms (Cyclotella and Stephanodiscus) in January and chlorophyta (Chlamydomonas, Carteria, Eudorina, Oocystis and Planktosphaeria) in February through April. The dinoflagellate Glenodinium also became numerous at this time and was dominant at all stations in March 1974.

Phytoplankton populations during May through December

were dominated by early summer and autumnal pulses of diatoms. Fragilaria was always dominant in May or June and was succeeded by a Cyclotella pulse in July. In 1972, samples were taken weekly and it was evident that the early summer Fragilaria and Cyclotella pulses originated in the inner bay and expanded outward in a wave-like manner on succeeding days throughout the middle and outer bays. Numbers of organisms declined outward from the inner bay. Navicula, on the other hand, was proportionately more numerous with increased distance from the inner bay and was usually the dominate organism in July or August at stations 4-6 in Boulder Basin. The blue-green algae, Anabaena, increased in numbers throughout July and August (except at Station 2 where Cyclotella remained dominant) and was the dominant organism in September after the early summer diatom pulses had declined. An autumnal pulse of Cyclotella always occurred throughout Las Vegas Bay and Boulder Basin in October associated with mixing. The autumnal diatom pulse lasted through December with numbers of Anabaena steadily declining.

The early summer pulse of Cyclotella has declined since 1972 and has been accompanied by an increase in the Anabaena population. There has also been an increase in the autumnal pulse of Cyclotella. In 1972, the early summer pulse of Cyclotella reached a maximum of over 53,000 cells/ml at

Station 2; there was also a well-defined pulse throughout Las Vegas Bay. In 1974 and 1975, the early summer pulse was much smaller (15,000 cell/ml) and numbers throughout Las Vegas Bay were lower. Total phytoplankton numbers have decreased since 1972, due mainly to the reduction in the early summer pulse of Cyclotella. Numbers of Cyclotella have increased during the autumnal pulse from 1500 in 1972 to 5000 cells/ml at Station 4 in 1975. The highest numbers of Cyclotella were found in October and November in 1974 and 1975, whereas, the highest numbers occurred in July 1972. Anabaena has become increasingly more numerous since 1972 in the outer bay and Boulder Basin. In 1975, Anabaena was dominant from July through September at Stations 4-6. Anabaena was dominant only in September in 1972 and 1974 following the early summer diatom pulses.

Phytoplankton Biomass - Figure 4 shows the seasonal variations in mean chlorophyll a and estimated cell volumes at Stations 2 and 6. Station 6 was not sampled in 1972. At Station 2 the highest single values for chlorophyll a (55.49 mg/m³) and cell volume (30.1 ml/m³) were found in April and May 1972, respectively. Both chlorophyll a and cell volumes indicate a reduction in the phytoplankton standing crop since 1972, although cell volumes in 1975 were higher than those found in 1974.

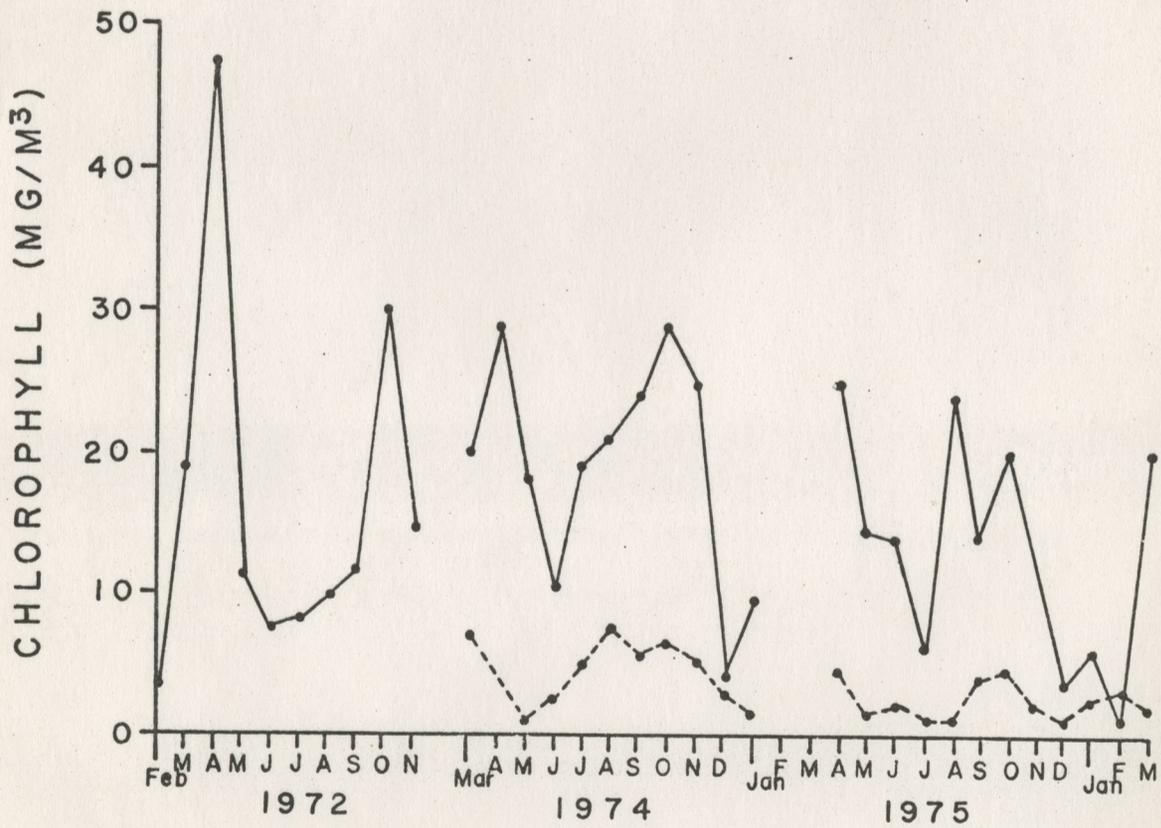
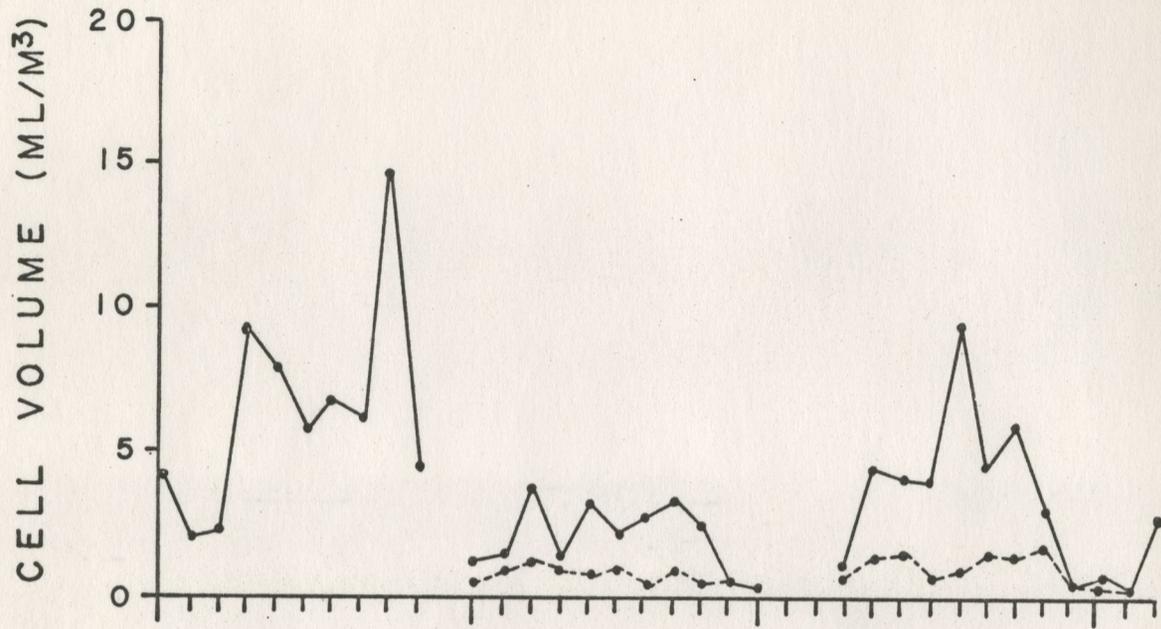


Figure 4. Mean Chlorophyll a and cell volumes at Stations 2 and 6. Station 2 (—), Station 6 (----).

Phytoplankton volumes were determined for only the dominant organisms or those organisms that were very large, therefore, total cell volumes were underestimated. Although the volumes were underestimated, they are useful as a comparative index. The cell volumes do reflect the higher eutrophic conditions at Station 2 due to nutrient enrichment. The higher cell volumes found in 1972 and 1975 were mainly due to Glenodinium and Fragilaria. Cell volumes in 1974 were low due to the complete absence of Fragilaria and lower numbers of Glenodinium. The early summer and autumnal pulses of Cyclotella had relatively little influence on biomass determinations; Cyclotella is small and it never comprised more than 15% of the total estimated biomass of the phytoplankton. Anabaena was more important, accounting for 15-50% of the total estimated biomass. Fragilaria, Glenodinium, Ceratium, and Oocystis were the dominant organisms on the basis of cell volumes.

There were two distinct chlorophyll a peaks. The first occurring in March and April when phytoplankton counts were low and dominated by chlorophyta. The second occurred in October and November during the autumnal Cyclotella pulse when chlorophyta numbers were declining. Chlorophyll a was

low in July when Cyclotella numbers were high. There was no evident relationship between Chlorophyll a and phytoplankton numbers.

Chlorophyll content per unit cell volume is dependent on a number of factors, such as species type, physiological state of the phytoplankton, and environmental conditions (Vollenweider, 1970). High cell volumes corresponded with high chlorophyll a only in the late summer and fall, a situation also described for Lake Erie (Munawar and Burns, 1976). The highest chlorophyll a concentrations per unit cell volume occurred in the spring (Table 5). The low chlorophyll a concentrations in the summer are possibly due to high light intensity, high water temperatures, and low nutrient concentrations at the surface.

Primary Production - Primary productivity was highest at Station 2 (Table 6) as were the other phytoplankton parameters. Generally, there was a reduction in productivity outward in Las Vegas Bay with the lowest productivity at Station 6 in Boulder Basin. Seasonally, the highest productivity occurred in August and September, ranging from 16,000 to 31,000 mgC/m²/day at Station 2 and from 3,000 to 8,000 mgC/m²/day at Station 6. Productivity generally increased from April through September with a substantial reduction in October. During the winter

Table 5. Mean Chlorophyll concentrations per unit cell volume (μg chlorophyll/ μl cell volume).

	1972	1974	1975	1976
Jan				9.41
Feb	7.90			3.58
Mar	11.09	16.08		7.33
Apr	24.88	17.38	20.99	
May	0.79	4.70	5.19	
June	0.91	8.86	3.80	
July	1.38	8.41	1.69	
Aug	1.42	10.51	2.54	
Sept	1.89	8.89	3.15	
Oct	2.05	9.45	3.51	
Nov	3.39	10.36	4.35	
Dec		7.20	7.87	

Table 6. Monthly primary productivity (mgC/m²/day) in Las Vegas Bay and Boulder Basin April 1974 through April 1976.

	1974 Station				1975 Station				1976 Station			
	2	3	4	6-8	2	3	4	6	2	3	4	6
Jan					1,652	1,158	1,207	783				
Feb					2,320	1,907			1,384	718	874	1,619
Mar						1,780	1,769		1,298	702	978	999
Apr	10,229	2,279	1,142	2,097	1,335	1,037	1,386		6,666	5,677	2,331	1,721
May	6,414	6,443	3,234	1,908								
June	7,742	3,394	4,100	3,135	9,442	4,180	1,843	1,500				
July	10,116	6,404	3,119	2,399	4,530	4,250	2,670					
Aug	16,793	12,862	7,310	5,988	16,663	14,843	7,642	7,386				
Sept	24,757	17,560	7,080	3,056	31,305	17,865	16,405	7,857				
Oct	5,339	4,827	5,844	2,678	7,600	8,800	4,242	4,048				
Nov	3,933	3,919	5,224	1,759								
Dec	2,160	1,551	2,066									

(December-February) productivity remained relatively high, ranging from 780 to 2300 $\text{mbC}/\text{m}^2/\text{day}$. Productivity was extremely high at all stations with means ranging from $8.3\text{gC}/\text{m}^2/\text{day}$ at Station 2 to $2.6\text{gC}/\text{m}^2/\text{day}$ at Station 6 (Table 7).

Nutrients - Phosphorous, nitrogen, and alkalinity concentrations in Las Vegas Wash, Las Vegas Bay, and Boulder Basin during April 1975 through February 1976 are presented in Appendix I. Table 8 summarizes total phosphorus and nitrite plus nitrate nitrogen (nitrate-N) concentrations for 1972, 1974, and 1975. In 1972, nitrate-N was determined only in September and October. Las Vegas Wash discharges approximately 800 kg phosphorus/day and 1100 kg nitrogen/day into Las Vegas Bay. There has been an increase in nitrate-N since 1974.

Total phosphorus levels were extremely variable in Las Vegas Bay and there was not an evident seasonal pattern. In Boulder Basin (Station 6), which was not directly influenced by Las Vegas Wash, total phosphorus ranged from approximately $22\ \mu\text{g}/\text{l}$ in December and January to $10\ \mu\text{g}/\text{l}$ in August and September. Vertically, total phosphorus was uniformly distributed or was slightly lower in the metalimnion. At Stations 2 and 3 higher levels were found in the metalimnion due to the density current from Las Vegas Wash. There was never a depletion in epilimnetic concentrations during summer stratification.

Table 7. Mean daily and total primary productivity in Las Vegas Bay and Boulder Basin, April 1974 through March 1975.

Station	Mean daily production g C/m ² /day	Mean yearly production g C/m ² /day
2	8.30	3030
3	5.34	1949
4	3.83	1398
6-8	2.65	965

Table 8. Mean phosphorus and nitrogen concentrations in Las Vegas Wash, Las Vegas Bay and Boulder Basin, 1972 through 1975.

Station	meters	1972 May-October		1974 March-December		1975 April-December		
		Total-P	Total-P	NO ₃	NO ₃ *	Total-P	NO ₃	NO ₃ *
1	0	4.236(.99)	4.156(.681)	8.54(2.86)		4.500(.240)	8.76(4.00)	
2	0	.049(.038)	0.302(.499)	0.15(.14)	0.05(.02)	0.800(.022)	0.19(.14)	0.09(.02)
3	0	.012(.006)	0.033(.008)	0.10(.09)	0.04(.02)	0.033(.009)	0.12(.08)	0.07(.06)
	30		0.091(.081)	0.41(.14)	0.45(.06)	0.107(.039)	0.42(.14)	0.51(.07)
4	0	0.10(.005)	0.027(.006)	0.11(.08)	0.03(.01)	0.017(.002)	0.13(.10)	0.04(.01)
	30		0.018(.008)	0.32(.09)	0.38(.07)	0.033(.018)	0.37(.10)	0.39(.04)
5	0		0.023(.007)	0.11(0.09)	0.03(.01)	0.016(.005)	0.13(.10)	0.04(.02)
	30		0.018(.008)	0.34(.09)	0.37(.07)	0.012(.004)	0.34(.06)	0.37(.02)
6	0		0.018(.003)	0.12(.09)	0.03(.02)	0.016(.004)	0.13(.10)	0.04(.01)
	30		0.014(.003)	0.35(.08)	0.37(.06)	0.013(.005)	0.34(.07)	0.37(.03)

Total P = Total phosphorus

NO₃ = Nitrite plus Nitrate nitrogen

Number in parenthesis = 1 standard deviation

* Mean values during summer stratification June - September

Surface nitrate-N at Station 6 ranged from approximately 300 $\mu\text{g}/\text{l}$ in January and February to 20 $\mu\text{g}/\text{l}$ during the summer. At all stations there was a depletion of epilimnetic nitrate-N during summer stratification (Table 8). Surface nitrate-N increased in October due to mixing which resulted in distribution of hypolimnetic nitrate-N throughout the water column.

Correlation Analysis - Correlation coefficients for phytoplankton parameters and nutrients (total phosphorus and nitrate-N) are presented in Table 9. *Cyclotella* was significantly correlated with total phosphorus and nitrate-N, and 57% of the variance in the *Cyclotella* population can be explained by total phosphorus and nitrate-N. *Anabaena* was negatively correlated with nitrate-N due to its occurrence only in the summer when nitrate-N was depleted from the epilimnion. Chlorophyll a and cell volumes were correlated only with total phosphorus. Primary productivity did not correlate with either total phosphorus or nitrate-N. Primary productivity was positively correlated with chlorophyll a and cell volume. Primary productivity and standing crop are usually not highly correlated and other investigators have reported inverse relationships between productivity rates and standing crop (Findenegg, 1965; Goldmen et al., 1968; Margalef, 1965; Verduin, 1959; Vollenweider and Nauwerck, 1961).

Table 9. Correlation coefficients for Cyclotella, Anabaena, Chlorophyll a, cell volume, and primary productivity.

Simple correlation coefficient
Variables

Dependent	Independent	n	r
Log ₁₀ Cyclotella #	Total phosphorus	32	.64*
Log ₁₀ Cyclotella #	Nitrate-n	32	.57*
Log ₁₀ Anabaena #	Total phosphorus	32	-.02
Log ₁₀ Anabaena #	Nitrate-n	32	-.61*
Chlorophyll <u>a</u>	Total phosphorus	48	.55*
Chlorophyll <u>a</u>	Nitrate-n	48	.18
Cell Volume	Total phosphorus	48	.30*
Cell volume	Nitrate-n	48	.02
Cell volume	Chlorophyll <u>a</u>	48	.66*
Primary Productivity	Total phosphorus	34	.09
Primary Productivity	Nitrate-n	34	-.20
Primary Productivity	Chlorophyll <u>a</u>	34	.56*
Primary Productivity	Cell volume	34	.37*

Multiple correlation coefficient
Variables

Dependent	Independent	n	R
Log ₁₀ Cyclotella #	Total phosphorus, nitrate-n	32	.76*

*significant at the .05 level

DISCUSSION

Early summer and autumnal pulses of diatoms are frequent occurrences in lakes (Hutchinson, 1967) and Cyclotella is a common organism demonstrating this pattern. Olsen (1975) found an early summer and autumnal pulse of Cyclotella in Canyon Lake, Arizona, which is similar to Lake Mead in climatic and chemical conditions, although it is much smaller. In Boulder Basin, the early summer Cyclotella pulse is restricted to Las Vegas Bay where there is an external nutrient source, Las Vegas Wash. The autumnal pulse is always associated with cooler water temperatures and mixing.

The dominance of Anabaena, a possible nitrogen fixer, during late summer would suggest that nitrogen is limiting at that time, although King (1970) and Shapiro (1973) have shown that this fact alone is not conclusive evidence of nitrogen limitation. The vertical distribution of nutrients involving summer depletion in the epilimnion further suggest that nitrogen may be the most important limiting nutrient. During summer stratification, nitrate-N is depleted from the epilimnion and significantly higher levels occur in the hypolimnion. Therefore, mixing results in increased nitrogen availability in the euphotic zone. This may be the most important factor behind the autumnal pulse of Cyclotella. Dissolved silica remained high (> 8 mg/l) throughout the

summer (U. S. G. S., 1975) and is, therefore, not a limiting factor. Phosphorus is uniformly distributed or shows a slight decrease below 10 meters and mixing does not result in higher phosphorus levels. If the autumnal pulse is due to higher levels of nutrients, nitrogen would appear to be the controlling factor. Nitrogen to phosphorus (N/P) ratios also show an excess of phosphorus (Table 10). The N/P ratios are significant because the ratio in living systems is about 8, therefore, when the N/P ratio exceeds 8 more nitrogen is present than can be utilized and, conversely, when the ratio falls below 8 there is an excessive amount of phosphorus (Verduin, 1967). The N/P ratios indicate that nitrogen is in short supply for most of the year in Las Vegas Bay and during the summer in Boulder Basin.

The increasing dominance of blue-green algae is usually associated with increased eutrophication (Edmonson et al., 1956; Fruh et al., 1966; Lund, 1969; Provasoli, 1969). The dominance of blue-green algae may be related to other factors, such as their freedom from grazing and their ability to withstand higher water temperatures and higher light intensity (Hutchinson, 1967; Lund, 1969; Golterman, 1975). In Las Vegas Bay and Boulder Basin, Anabaena has become increasingly more dominant since 1972 and was completely dominant throughout the summer (July-September) at Stations 3-6 in

Table 10. Nitrogen (Nitrate-N and Ammonia-N) to phosphorus (total phosphorus) ratios for Las Vegas Bay, Stations 2-4 and Boulder Basin, Station 6, 1975.

	Stations			
	2	3	4	6
April	1.6	4.5	15.9	14.7
May	1.5	9.0	13.3	13.8
June	2.1	2.3	6.1	12.0
July	2.6	2.0	3.5	3.8
Aug	1.8	1.8	3.3	5.0
Sept	2.4	1.9	3.3	3.7
Oct	2.2	2.4	4.5	5.5
Nov	9.1	6.7	11.0	11.5
Dec	10.0	12.0	15.8	13.6

1975. The complete dominance of Anabaena in 1975 may be related to a flash flood in July which discharged a large quantity of nutrient rich sediments from the Las Vegas Wash marsh system into Las Vegas Bay. The increasing dominance of Anabaena would suggest increasing eutrophic conditions in Boulder Basin.

Phosphorus loading from Las Vegas Wash has increased since 1972 from a mean daily rate of 525 kg/day to 792 kg/day in 1975. The phosphorus loading from Las Vegas Wash is extremely high when considering the Colorado River, which has 200 times the volume and discharges only 950 kg/day into Lake Mead. Total phosphorus loading for Boulder Basin ($1.944 \text{ g/m}^2/\text{yr}$) is in the "dangerous" level proposed by Vollenweider (in press). A reduction in the phosphorus loading from Las Vegas Wash to 360 kg/day would result in a "permissible" loading rate in Boulder Basin. Nitrogen loading from Las Vegas Wash in 1974 and 1975 was approximately 1100 kg/day. Nitrogen concentrations were not determined for Las Vegas Wash in 1972.

Although phosphorus loading has increased, phytoplankton numbers and biomass have decreased since 1972. This may be directly related to lake elevations. In 1972, the water level was about 6 meters lower than it was in 1974 and 1975. The higher water levels increased the volume of the inner Las Vegas Bay alone by approximately $50 \times 10^6 \text{ m}^3$. The increased volume

would result in a greater dilution of the Las Vegas Wash influent, thereby decreasing the availability of the nutrients to the phytoplankton.

The trophic classification of Lake Mead has received considerable attention in the past few years. Phytoplankton species indices have been discussed in the 1975 report and by Staker et al. (1974). These indices have given results ranging from polluted eutrophic to mesotrophic or even oligotrophic conditions. Everett (1972), based on Rodhe's (1969) primary productivity categories, classified Boulder Basin as a polluted, eutrophic body of water. Rodhe's classification is based on temperate lakes and does not take into account the extended growing season in a subtropical lake and, therefore, is probably not appropriate to conditions in Lake Mead. Our primary productivity estimates are similar to Everett's and, in addition, show very high production in Las Vegas Bay. Primary productivity in Boulder Basin is comparable to other tropical or subtropical lakes (Talling, 1965; Berman and Pollinger, 1974; Melack and Kilham, 1974) and conditions in Boulder Basin are, therefore, probably not as serious as Everett stated. Vollenweider (1970) proposed the following classification based on maximum phytoplankton cell volumes:

Ultra-oligotrophic	1 ml/m ³
Mesotrophic	3 to 5 ml/m ³
Highly eutrophic	10 ml/m ³

Using this classification, Las Vegas Bay is highly eutrophic with the rest of Boulder Basin being in the mesotrophic range.

Zooplankton Community
of Boulder Basin

by

Thomas Burke

INTRODUCTION

Preliminary investigations in 1974 showed peak zooplankton concentrations within the metalimnion of Boulder Basin, Lake Mead. A similar circumstance had previously been noted by Shapiro (1960), who concluded that zooplankton were a major factor in the metalimnetic oxygen depletion of Lake Washington. This report is the result of a one year study of the zooplankton community of Lake Mead. Its purpose was to determine if any unique differences existed between the zooplankton community of Lake Mead and other limnetic communities.

Seasonal succession, species composition, vertical distribution, and specific depth affinities of the zooplankton community were examined. The data were compared to those of other investigators wherever possible.

METHODS AND MATERIALS

Zooplankton samples were collected at approximately two week intervals from June through October and at monthly intervals for the remainder of the year. Station 5 was selected as the permanent sampling location for these collections. This station is located in close proximity to the intakes of the Alfred Merrit Smith Water Treatment Plant. The water depth increases sharply from the shore

to a depth of 65 meters. This allowed for very little contamination of the sample with littoral organisms. Easy access to and from Lake Mead Marina was also favorable to the selection of this station, as many samples were taken over a 24-hour period.

Samples were taken at five meter intervals from surface to 45 meters with a portable water pump. Tonolli (1971) recommended this type of device where large numbers of samples need to be taken in a short period of time. The pump type sampler has been shown to successfully minimize damage to plankton samples used for identification purposes (Aron, 1958). It was also shown to have zooplankton fishing abilities equal to a conventional plankton tow net (Icanberry and Richardson, 1973).

Summer samples were collected with a Sears Model #563.2692 D-C powered pump with an average flow-through velocity of 8 liters/minute. The winter samples were taken with a 1.5 horsepower gasoline-driven pump with an average velocity of 12.5 liter/minute. Both pumps were attached to a heavy-duty, reinforced rubber hose. The bottom of the hose was weighted with a plexiglass plate designed to keep the hose vertical and to allow an even draw from a narrow, horizontal band. From each depth, forty liters of lake water were filtered through a #20 mesh, nylon, plankton net (76 μ . diameter pore). Samples were immediately preserved in a 5% formalin solution as recommended by Lackey (1938).

Counting and identification of the plankton was done on a Wetzlar Model #600112 compound stereo microscope. Taxonomic identifications were based on Coker, (1943); Davis, (1955); Edmonson, (1959); Gurney, (1931, 1933); Marsh, (1929); and Pennak, (1953). All organisms were identified to genus, and to species when possible. One milliliter subsamples were placed in an open Sedgewick Rafter counting cell. All organisms in five such aliquots were tallied. This represented 5 to 10% of the summer samples and 10 to 20% of the winter samples. With the exception of eggs, organisms less than 50 microns in diameter were not counted (i.e. ciliates and zoo-flagellates).

RESULTS AND DISCUSSION

The limnetic zooplankton community of Boulder Basin was dominated by Rotatoria, Cladocera, and Copepoda. Other organisms such as ciliates, zoo-flagellates, insect larvae, and water-mites were often found in the samples, but their numbers were usually very low. One species of Diffflugia, a protozoan, was very abundant in the 8 May samples. The sample was accidentally discarded and verification of the counts cannot be made. Except for a small number of Diffflugia in the mid-July sample, these organisms have not reappeared.

Rotifers - The rotifers are represented by twelve genera, five of which were classified as major components of the community, and are listed in Table 11. While one or more of the following seven genera were often present in the samples, no single genera ever represented more than 5% of the rotifer population:

Trichocerca, Notholca, Platylabus, Pleosoma, Lecane, Monostyla, and Brachionus.

Cladocera - Three genera of cladocerans were identified from our samples. The first two, Daphnia and Bosmina, were considered to be truly limnetic organisms, but the third genus, Alona, is more typically a littoral organism. (Pennak, 1953). All Daphnia were grouped together, but tentative identification indicates three species may be present. For Bosmina, only one species was found, B. longirostris.

Copepoda - The limnetic copepods are represented by four species, two in the order Cyclopoida (Cyclops bicuspidatus thomasi and Mesocyclops edax) and two in the order Calanoida (Diaptomus clavipes and Diaptomus siciloides).

TABLE 11. Major Rotifers in the Water Column at Station 5 ($\#/m^2 \times 10^3$).

	May 28	Jun 16	Jul 1	Jul 17	Aug 19	Sept 17	Oct 15	Nov 19	Dec 23	Jan 29	Feb 19	Mar 24	Apr 27
<u>Asplanchna</u>	50	320	98	12	0	0	0	2	20	150	35	0	5
<u>Syncheata</u>	0	2	106	80	3	45	70	630	365	5000	180	3	0
<u>Polyarthra</u>	260	75	92	96	19	41	65	43	16	340	8	5	150
<u>Collotheca</u>	0	25	240	27	15	25	0	15	18	20	13	40	16
<u>Keratella*</u>	821	159	53	94	9	5	11	11	64	13	3	10	50
<u>All Others</u>	10	10	25	18	0	8	3	25	0	5	0	0	0
TOTAL	1120	591	614	327	46	124	149	726	483	5528	239	58	221**

* Keratella-- Total of K. quadrata and K. cochlearis.

Seasonal Succession

A review of the literature indicates that zooplankton may have one, two, or three seasonal periods of abundance. These high and low periods may be caused by physical, chemical, and/or biological conditions, with a combination of these usually occurring. Single species show different cycles from lake to lake and may even show differences from year to year in the same lake. These seasonal variations seem to change the community make-up at the species level only. Pennak (1957) found that at any given time a typical zooplankton community is dominated by one copepod, one cladoceran, and two to four numerically dominant rotifers.

The zooplankton of Lake Mead show varying patterns of seasonal succession. Table 12 shows the number of organisms of each major group occurring in the samples from May 1975 through April 1976. The population of each group shows relatively high numbers of individuals during June/July, October/November, and January/February. Although three peak periods occur in the lake, analysis of the community by each species shows that the majority of the organisms are either dicyclic or monocyclic.

As mentioned earlier, the rotifer population of Boulder Basin was best represented by five genera, each of which was the dominant rotifer on one or more of the dates sampled (Table 11). Not only were different species dominant during

Table 12. Number of organisms ($\#/m^2 \times 10^3$) in each major group in the upper 25 meters of the water column.

	May 8	Jun 16	Jul 17	Aug 19	Sept 17	Oct 15	Nov 19	Dec 23	Jan 29	Feb 19	Mar 24	Apr 27
Total Copepods (CI-CZ Adult)	259	861	766	817	351	905	944	177	172	359	345	576
Copepod Naoplii	952	1220	2150	1430	211	200	1040	195	318	1307	830	1250
Total Cladocera	25	192	46	106	160	63	491	376	162	464	226	140
Total Rotifera	1120	591	327	46	124	149	726	483	5528	239	58	221
Total	2356	2864	3289	2399	846	1317	3207	1231	6180	2364	1459	2187*

the peak periods, but the early summer period was marked by different species gaining or losing dominance within this period.

Asplanchna was dicyclic, being abundant in June and January. The number of organisms in the water column was 320,000 and 150,000 per square meter of surface area, respectively. The counts for January may be conservative due to the presence of Syncheata, which was very abundant. Syncheata was present in all stages of development from newly hatched individuals to large sac-like adults. This latter form is very similar to a small saccate form of Asplanchna. The majority of Asplanchna were of a very large campanulate form (twice the size of adult Syncheata), but a qualitative re-examination of January samples showed a small number of saccate Asplanchna to be present. It is interesting to note that the June population also contained a large campanulate form of Asplanchna. Gilbert (1973) reports that Asplanchna sieboldi will greatly increase in size in the presence of any large prey species. High numbers of Syncheata in January and of Keratella and Polyarthra in May and June represent an adequate food for the Asplanchna and may have given rise to this large form.

Syncheata was abundant in January and reached a density of 320 organisms/liter at the ten meter depth for this date. While this seems to be a very high figure, Pennak (1953)

reports finding single rotifer species at densities in excess of 500/liter. Syncheata was also abundant in July and November, suggesting that the species may be tricyclic. However, the November peak results from hatching of resting stage eggs. This generation then goes on to reproduce amictically, reaching its peak in January. Therefore the apparent tricyclic occurrence of Syncheata is actually dicyclic. The phenomenon of diapause or resting stages will be discussed in detail in a later section of this paper.

Two species of Keratella were identified from our samples. Keratella cochlearis was the dominant species, being present in all of the samples and responsible for the peaks in May and December. Keratella quadrata was present in lower numbers, and was found only during periods of peak zooplankton abundance (June/July, October, and January).

Collotheca seems to be dicyclic, having peaks in March and July. Polyarthra also appears to be dicyclic, although its peaks occur in January and May. It is difficult to determine if the fluctuations in the Polyarthra population between June and December are truly cyclic responses. A third peak may have occurred in October, although numbers were much lower than those found in January and May.

Of the five genera of rotifers discussed, none seem to have both of their dominant periods in the same two months. There was some overlap in mid-winter but the late spring or early summer populations were spread over a three-month period (Table 13).

The two limnetic cladocerans were monocyclic with their peak populations being out-of-phase (Table 14). Daphnia had its maximum abundance in June and July, while Bosmina was most abundant in November, December, and February. It is common practice when counting organisms for quantitative analysis to neglect fragments or incomplete specimens. Ehippial stages of Daphnia were extremely abundant in the surface film starting in February and continuing into May. This stage contains resting eggs attached to the valve or carapace of the adult. The valve separates from the head of the organism at the next molt. The ehippia then float to the surface and concentrate in windrows as a result of surface winds and Langmuir circulation. This makes it difficult to get a quantitative estimate of their numbers. It was noted that zooplankton samples from March and April contained a large number of Daphnia heads. Assuming that each head represented an adult Daphnia that had given rise to an ehippium, one could add these counts to the population

Table 13. Periods of peak abundance of the major rotifers
at Station 5, Boulder Basin

<u>Organism</u>	<u>Peak Months</u>	
<u>Asplanchna</u>	June	January
<u>Syncheata</u>	July	January
<u>Polyarthra</u>	May	January
<u>Keratella</u>	May	December
<u>Collotheca</u>	July	March

Table 14. Cladocerans in the upper 25 meters
(#/m²X10³).

Organism	Sample Date						
	May 8	Jun 16	Jul 1	Jul 17	Jul 31	Aug 19	Aug 27
<u>Daphnia</u>	16	190	260	7	71	32	9
<u>Bosmina</u>	17	2	1	39	105	74	151
Total	25	192	261	46	176	106	160

of water column, Station 5

Sept 17	Oct 15	Nov 19	Dec 23	Jan 29	Feb 19	Mar 24	Apr 27
0	0	2	0	14	6	48	121
160	63	489	376	148	458	178	19
<hr/> 160	<hr/> 63	<hr/> 491	<hr/> 376	<hr/> 162	<hr/> 464	<hr/> 226	<hr/> 140

and possibly obtain a better estimate of the actual population density of Daphnia. There were 67,000 heads/meter² in the water column in March. The total Daphnia in the water column was estimated as 48,000 individuals/meter². Summing these two figures, the total Daphnia population for March appears closer to 115,000 individuals/meter². For April, the heads totaled 112,500/meter² and the Daphnia totaled 121,000/meter². This would give an adjusted population estimate of 233,500 individuals/meter². These adjusted population estimates for Daphnia appear to be more accurate than those listed for March and April (Table 14).

The copepod population had three periods of abundance. The summer period was marked by three months of relatively high numbers (Table 15). This was due to an exchange of dominance between the two cyclopoid copepods. The other two dominant periods occurred in October/November and March/April. All three periods had a different species composition.

The predaceous cyclopoid, Cyclops bicuspidatus thomasi, (McQueen, 1969) was monocyclic, remaining dominant from February into July. During the month of July it was replaced by another predatory species, Mesocyclops edax (Confer, 1971). M. edax remained the dominant cyclopoid from July through December.

Table 15. Copepods in the upper 25 meters of the water column at Station 5, Boulder Basin ($\#/m^2 \times 10^3$).

SPECIES	Jun 16	Jul 1	Jul 17	Jul 31	Aug 19	Aug 27	Sept 17	Oct 15	Nov 19	Dec 23	Jan 29	Feb 19	Mar 24	Apr 27*
<u>Cyclops bicuspidatus thomasi</u>	106	170	280	158	88	25	11	45	12	21	13	106	130	110
<u>Mesocyclops edax</u>	3	11	42	412	306	220	143	326	170	81	9	43	15	9
<u>Cyclopoid copepodids</u>	658	225	386	472	341	415	108	418	640	64	104	160	220	480
<u>Diaptomus clavipes</u>	22*	50*	4	27*	14	2	3	18	15	2	1	3	8	0
<u>Diaptomus siciloides</u>			9		3	2	9	56	90	1	9	19	43	21
<u>Diaptomid copepodids</u>	81	40	45	22	65	76	77	42	29	8	37	28	29	75
<u>Copepod nauplii</u>	1220	993	2150	1360	1430	843	211	200	1040	195	318	1307	830	1250

* Both species counted together

The two species of Diaptomus, D. clavipes and D. siciloides, never obtained the numerical dominance of the cyclopoids. D. siciloides reached its highest population in November, when it represented over 30% of the adult copepod community. This was more than five times the numbers reached by either Cyclops bicuspidatus thomasi or D. clavipes.

D. siciloides was monocyclic, first appearing in July and reaching its peak population in November. The samples for 16 June, and 31 July will be re-examined to determine what per cent of the Diaptomus population was actually D. siciloides. This will also allow for a better interpretation of the D. clavipes population, which at this time appears to have no real period of abundance.

In general, copepods have six nauplii instars, five juvenile instars (copepodids), and a single adult stage. The Diaptomus nauplii and the Cyclopoid nauplii were grouped together in our counts. The adults listed in Table 15 represent the combination of both the fifth copepodids and adults for each species. The groups listed as copepodids were the total of the first four juvenile instars.

Vertical Migration

Diurnal vertical migration of zooplankton is a well-known and well-documented phenomenon. Hutchinson (1967) describes three types of vertical migration: (1) nocturnal migration,

with a single maximum in the upper stratum at night, (2) twilight migration, with a maximum at dawn and dusk, and (3) reverse migration, with one maximum at the surface at mid-day. Possible explanations for the causes of the migrations include light, temperature, pH, conductivity, and predator avoidance. Pennak (1944) concluded light to be the single most important factor.

The vertical range of the migration varies from species to species and from lake to lake. Campbell (1941) found that physical and chemical factors in Douglas Lake were able to restrict the range of migration. He noticed that as the zone of hypolimnetic oxygen depletion increased during the summer the rotifer population was constricted into the upper 15 meters of the water column.

The rotifer population of Boulder Basin undergoes vertical migration during both thermally stratified and unstratified periods. Staker (1974) found the average range of Keratella cochlearis and Polyarthra in Boulder Basin during January to be 4.8 and 2.6 meters, respectively. These amplitudes of vertical migration agree with those discussed by Pennak (1953). He states that amplitudes of one to three meters are common, with amplitudes of eight to ten meters being very unusual. George and Fernando (1970) disagree with Pennak's average. Their report

covered a one-year period and showed the vertical amplitudes to vary between summer and winter. The data they present shows Polyarthra vulgaris to migrate 1.8-4.8 meters in February, 3.0-4.1 meters in April, 7.0-10.0 meters in June, 3.0-8.6 meters in July, and 4.8-8.4 meters in August.

The Lake Mead population of Polyarthra shows similar patterns to those listed above. Figure 5 shows the vertical profiles of Polyarthra for three consecutive samples. The vertical amplitudes in May and June were about 5-10 meters and similar to the results reported by George and Fernando (1970). In July, when thermal stratification was well established and an oxygen minimum was present in the metalimnion, Polyarthra was restricted to the upper 20 meters, possibly because of the low oxygen concentrations.

Thermal stratification appears to have an effect on the vertical distribution of other rotifers in Boulder Basin (Fig. 6). Both winter and summer Asplanchna populations had high concentrations in the upper ten meters of the water column. The summer population was more or less restricted to this region, but the winter population extended down to a depth of 35 meters. Collotheca also showed a different profile for winter and summer. While summer populations of Collotheca showed highest concentrations in the top ten meters, it was rare to find any of this genera above ten meters

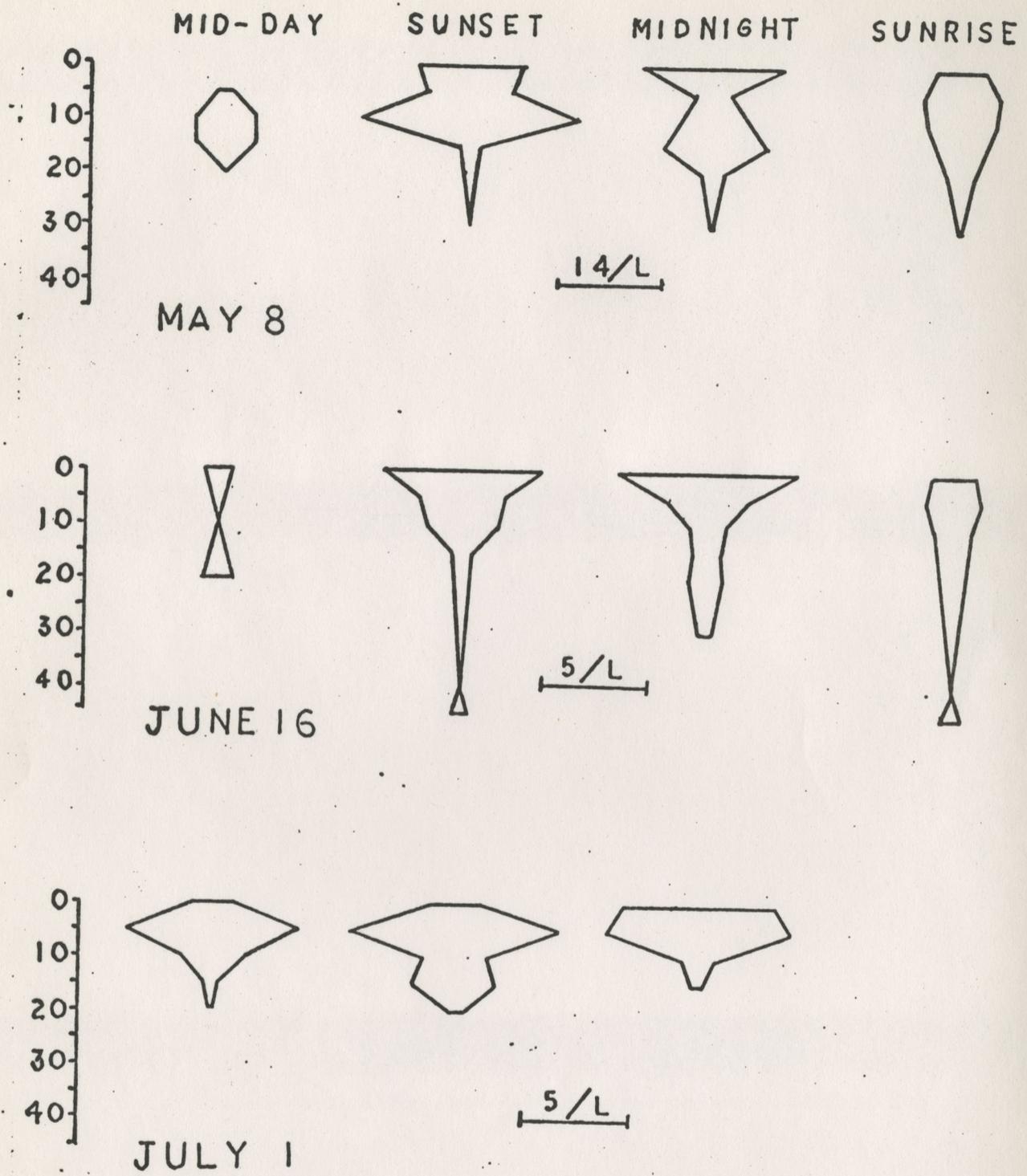
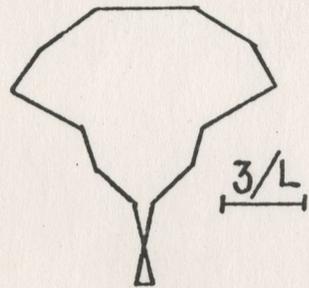
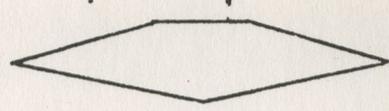
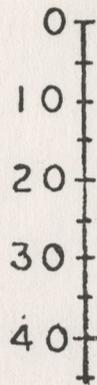


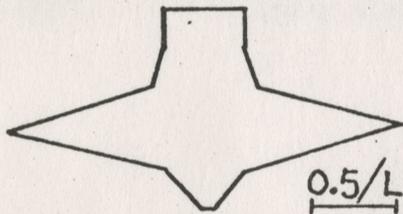
Figure 5. Vertical distribution of *Polygrthra* at Station 5, Boulder Basin (depths in meters).

ASPLANCHNA

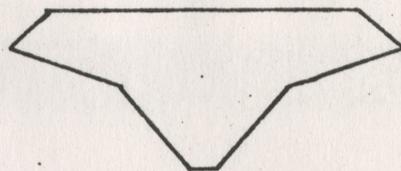
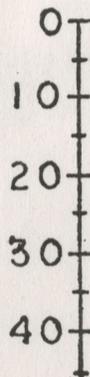
JANUARY



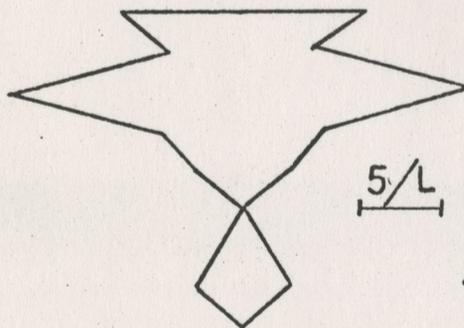
JULY

 $3/L$ COLLOTHECA

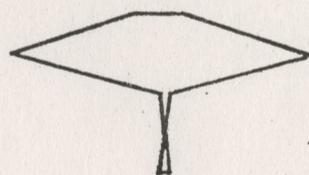
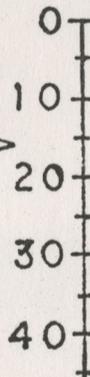
JANUARY



JULY

 $4/L$ SYNCHEATA

DECEMBER



JULY

 $5/L$

Figure 6. Vertical distribution of winter and summer populations of three rotifers at Station 5, Boulder Basin, sunset samples (depths in meters).

during the winter. This rotifer is encased in a mucilaginous sheath and is considered to be a poor swimmer. Winter populations appeared to have a considerable amount of detritus and particulate matter adhered to this sheath, more so than that found on the summer organisms. Syncheata showed vertical distribution similar to that of Asplanchna. Organisms were concentrated in the top 10 meters, extending down to 20 meters in the summer. Winter populations ranged from 5 to 45 meters, with the concentrations being highest at 15 meters.

Vertical profiles for the cladocerans are shown in Figure 7. Accurate values for the vertical amplitude are difficult to obtain with samples taken at five-meter intervals. Staker (1974) reported that Bosmina migrated 4.7 meters. While Figure 7 shows some possible migration, the bulk of the population remains between 10 and 20 meters. The sunrise sample indicates that the bulk of the population may have been between sampling depths. Both cladocerans showed differences in summer and winter profiles (Fig. 8) similar to the rotifers.

The vertical amplitudes of migration for the copepods is generally believed to be much greater than that of the rotifers and about equal to that of the cladocerans. Staker (1974) concluded that the copepods of Boulder Basin did not

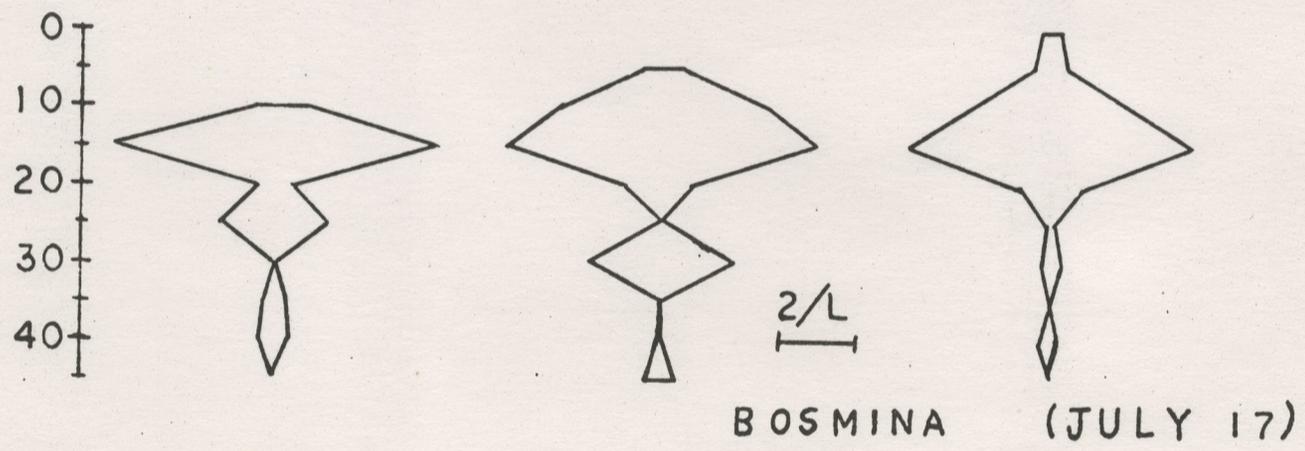
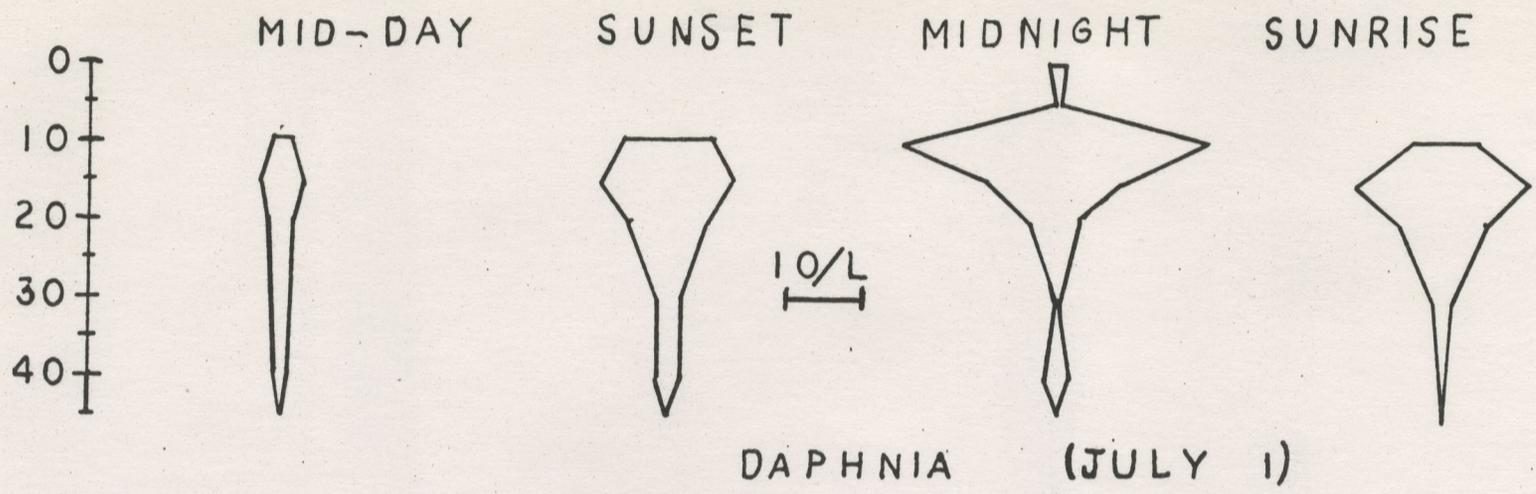


Figure 7. Vertical migration of Cladocerans at Station 5, Boulder Basin (depth in meters).

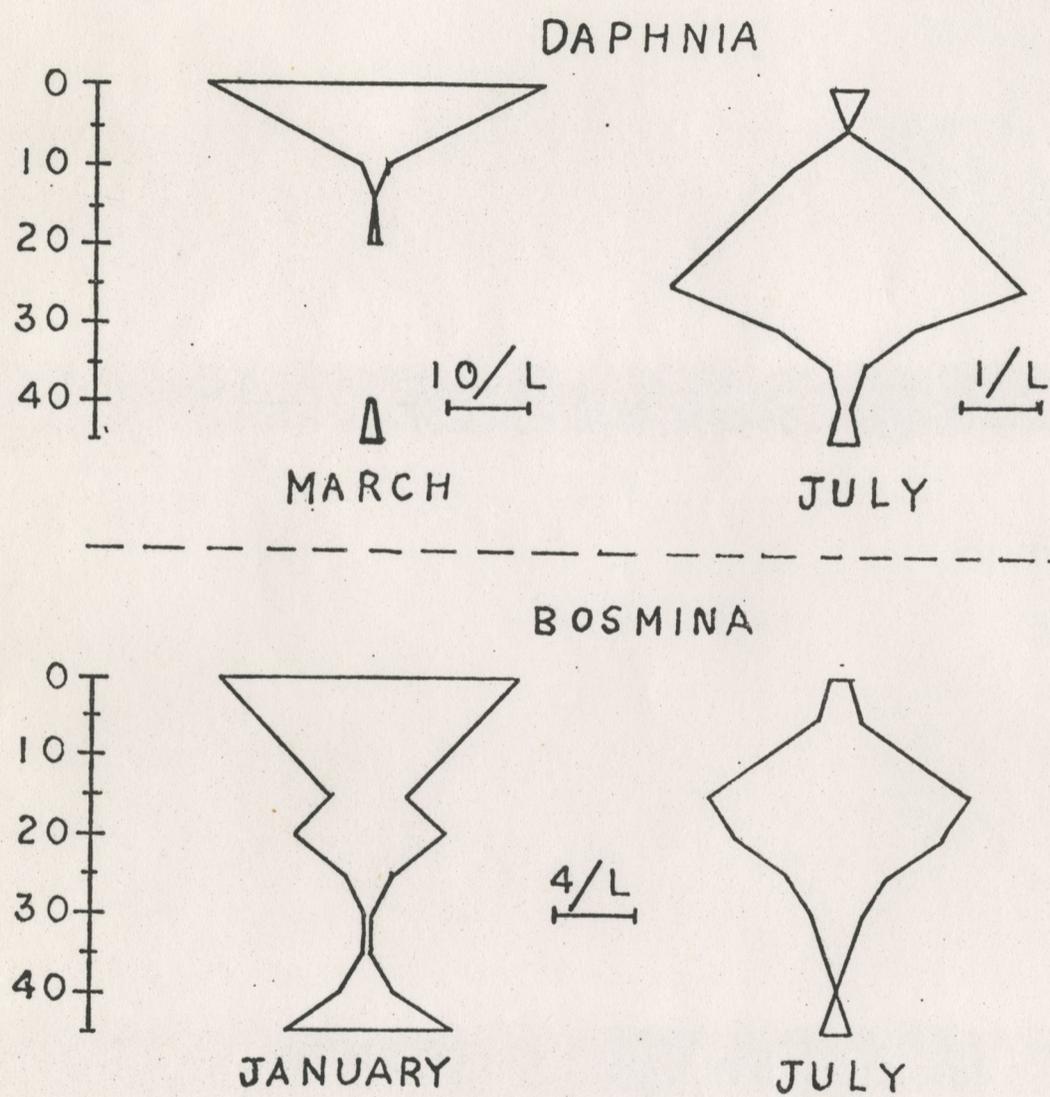


Figure 8. Vertical distribution winter and summer Cladoceron populations, sunset samples, Station 5, Boulder Basin (depth in meters).

migrate during the winter. His data did not report any breakdown of instars of these copepods. Marshall and Orr (1955) report that the juvenile instars of the marine copepod, Calanus finmarchicus, showed different vertical ranges and patterns from those of the adults for both winter and summer populations. This was evident in the summer populations of copepods from Lake Mead. A comparison of copepod nauplii, cyclopoid copepodids I-IV, and C. b. thomasi V & IV from early July showed the vertical migration to be increasing with each group, respectively, with the nauplii undergoing the least amount of vertical change (Fig. 9).

Data from the 31 July sample shows a different pattern. C. b. thomasi had its highest density for the sunset sample at 15 meters. Mesocyclops edax had its greatest concentrations at the surface at this time. The cyclopoid copepodids showed a peak at each of these depths, indicating different patterns for these two species (Table 16).

Diaptomus populations were generally low, but a comparison of the winter and summer profiles indicated that a more confined distribution exists during thermal stratification (Fig. 10). Comita and Anderson (1959) report a similar observation. They found more diaptomids in the epilimnion than in the hypolimnion of Lake Washington during times of thermal stratification.

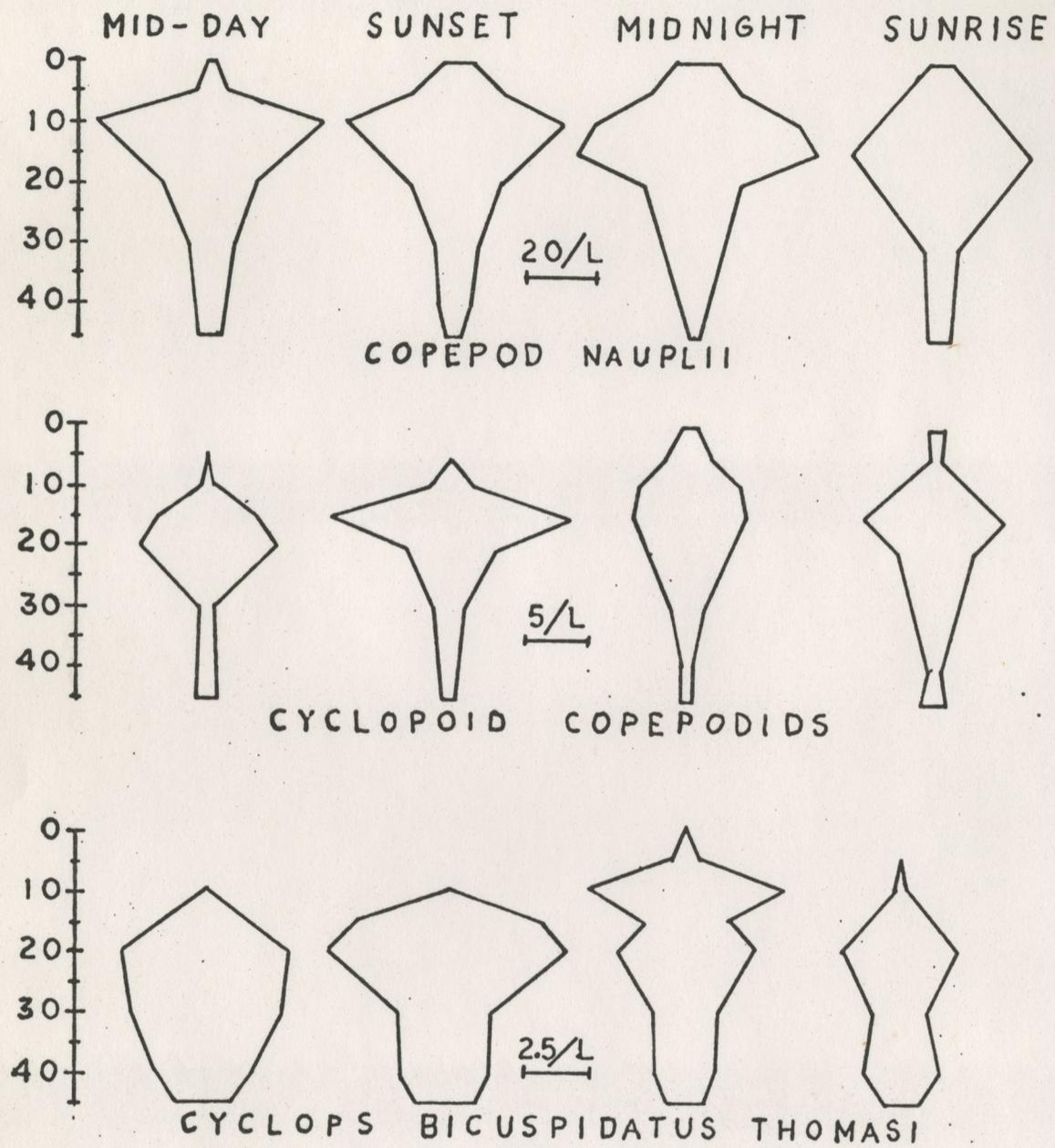


Figure 9. Vertical profiles of copepod nauplii, cyclopoid copepodids, and Cyclops bicuspidatus thomasi at Station 5, Boulder Basin, July 1975 (depth in meters).

Table 16. Cyclopoid copepodes (#/l) at Station 5, Boulder Basin, sunset samples.

Depth (m)	Sample Date										
	7-1	7-31	8-27	9-17	10-15	11-19	12-23	1-29	2-19	3-24	
<u>Cyclops</u>	0	0	0	0	0	0	1.8	0	5.6	5.1	
<u>bicuspidatus</u>	5	0	0	0	0.3	0.2	1.0	0.9	3.1	8.0	
<u>thomasi</u>	10	0	1.6	0	0.5	0	0.5	0.2	2.7	1.4	
	15	7.5	19.9	0.4	0	1.8	0	0	0.9	1.7	
	20	9.9	10.2	2.4	0.3	2.4	0.2	0	1.7	1.8	
	25	-	6.5	0.7	0.9	0.5	0	0	1.2	2.0	
	30	2.6	2.6	1.1	0.3	3.0	0	0	2.1	1.5	
	35	-	-	0.8	0	0	0.4	0.1	1.0	1.4	
	40	3.4	-	0	0.7	0	1.0	0.1	0.7	1.6	
	45	2.6	-	0	0	0.5	0.5	0.2	0.6	2.1	
<u>Meso-</u>											
<u>cyclops</u>	0	0	19.2	1.7	0	0.2	0	0.1	2.6	0.8	
<u>edax</u>	5	0	13.5	5.9	1.8	13.2	0.2	0	2.0	1.3	
	10	0	15.6	8.0	4.9	9.2	0.4	0	2.2	0	
	15	1.4	3.4	8.5	6.3	13.6	0.9	0	0.6	0.2	
	20	0.5	1.2	8.4	3.7	5.4	2.8	0.1	0	0.2	
	25	-	5.4	3.9	2.0	4.9	4.4	0	0	0.5	
	30	0	2.8	2.5	2.6	5.3	9.4	0.4	0	0	
	35	-	-	1.6	3.3	3.0	8.8	1.4	0.1	0.2	
	40	0	-	0.8	7.6	7.9	5.2	3.2	0.4	0.1	
	45	0	-	1.6	2.3	2.7	1.9	11.2	0.3	0	
<u>Cyclopoid</u>											
<u>copepodids</u>	0	0	21.7	5.6	1.6	0	1.8	1.7	1.4	7.7	12.7
	5	0	6.9	4.7	1.2	19.2	2.0	1.1	3.3	10.9	11.6
	10	4.8	11.1	12.4	2.4	16.0	6.1	0.4	3.7	10.8	5.1
	15	20.2	20.4	24.7	2.5	25.2	11.7	0.3	1.0	0.8	2.6
	20	7.4	12.3	17.4	7.7	12.2	16.6	0.2	2.1	0.6	3.4
	25	-	10.8	9.7	2.7	6.2	20.2	0.5	1.9	0.2	4.2
	30	2.6	6.0	5.0	2.7	1.9	26.8	0.4	2.6	0.6	2.7
	35	-	-	2.0	0.8	2.4	28.2	0.8	0.7	0	1.8
	40	1.9	-	0.4	0	0	13.0	1.3	0.7	0.1	1.5
	45	1.5	-	0.9	0	0.5	2.9	2.4	1.7	0.3	2.5

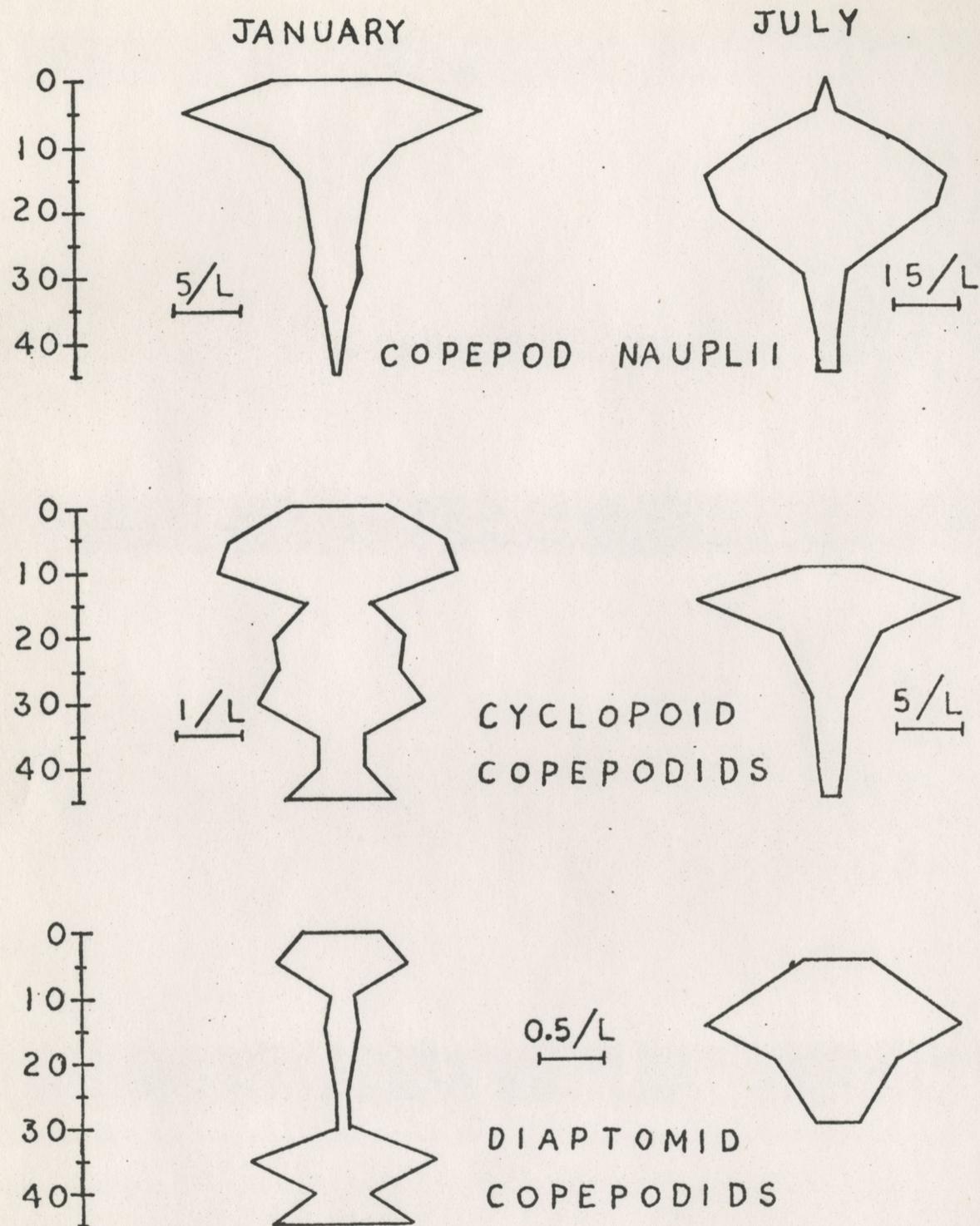


Figure 10. Vertical distribution of early copepod stage winter and summer populations, sunset samples, Station 5, Boulder Basin (depth in meters).

Depth Affinities

The previous illustrations were used to depict vertical distribution relative to vertical migrations, but they also illustrate the preference of these organisms for specific zones of the water column. The rotifers showed summer concentrations restricted to the upper 20 meters. The cladocerans and copepods showed a preference for the 10 to 25 meter layer during summer stratification. Marshall and Orr (1955) attempted to explain this preference as it applies to copepods which migrate into and through this layer. They suggest that vertical migration is a means by which the animal can sample fresh layers of water, and these organisms may be able to stop at layers where optimal conditions exist (e.g., food and light).

In general, the herbivorous members of the zooplankton community show a definite affinity for the metalimnion during summer stratification (Fig. 11). These data show that the zooplankton do maintain a substantial population within the metalimnion during thermal stratification.

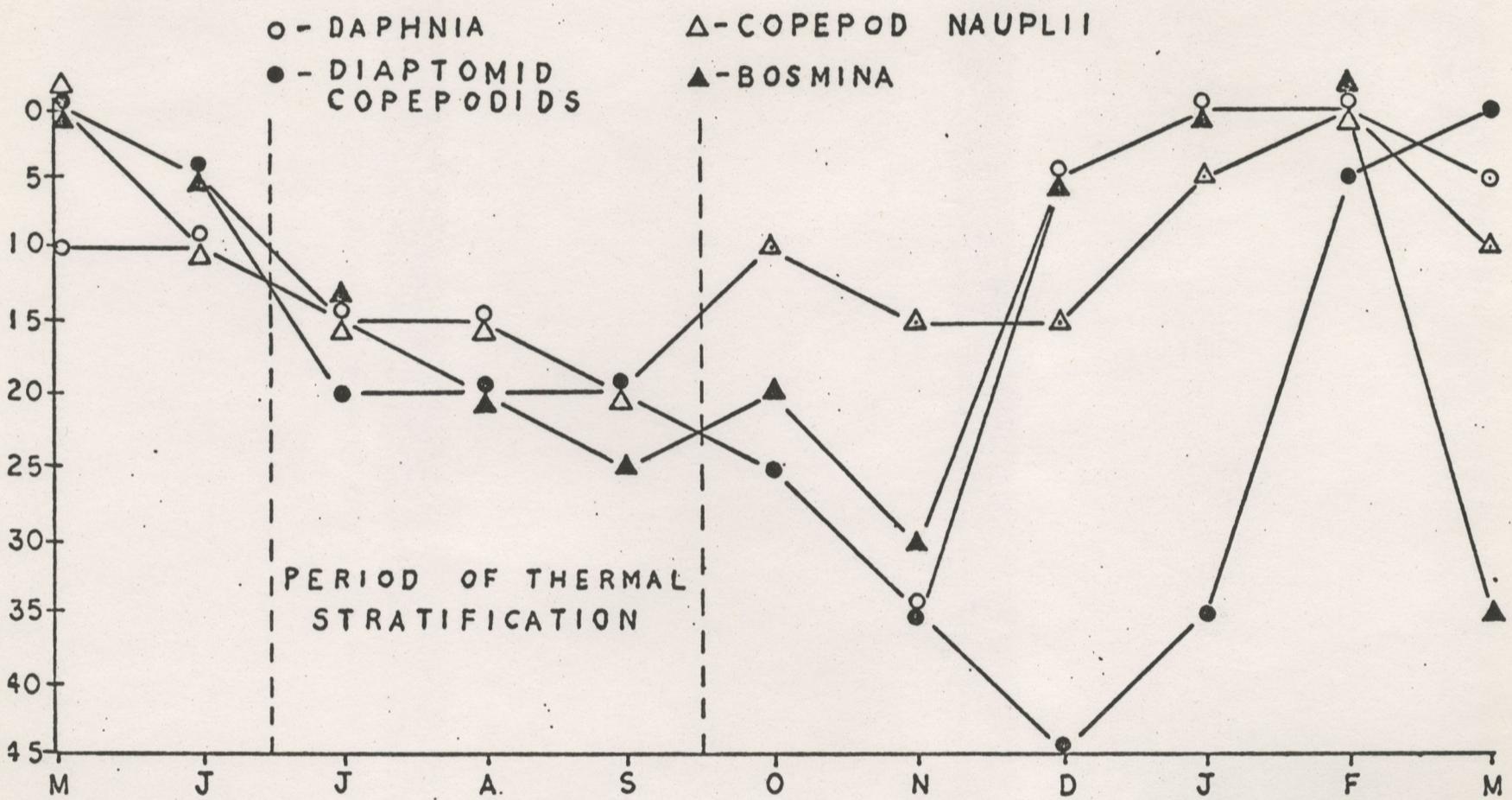


Figure 11. Depth of maximum density for zooplankton grazers at Station 5, Boulder Basin, May 1975 to March 1976 (depth in meters).

Diapause

Brief mention of resting stages has been made in this report. The occurrence of this phenomenon is known for a variety of common freshwater zooplankters. Some organisms develop resting eggs, while others may have cyst-like stages similar to pupal stages of insects. The ability of an aquatic organism to exist in a dormant state is generally considered to be a successful adaptation to its environment. This would allow the species to withstand periods of stress such as summer drought, anoxic water conditions, and cold winter periods of low food availability. Species from each major group of zooplankton from Boulder Basin have been found to exhibit some type of resting stage.

In the rotifers both Syncheata and Polyarthra were found hatching from resting stage eggs. Resting eggs of Syncheata first appeared in the October 30 samples, with higher numbers being found in the November 19 samples. Vertical oxygen profiles showed that rapid mixing occurred during the October/November period. The water column was completely mixed from surface to 16 meters on 1 October, to 20 meters on 15 October, to 29 meters on 30 October and to 40 meters on 19 November. This mixing resuspended these eggs into the water column where they developed into the adult forms. The resting egg is different from the summer egg of Syncheata, having a thicker outer

wall and numerous spines. Pennak (1953) gives diagrams of common rotifer resting eggs, illustrating these characteristics.

Polyarthra first indicated the presence of a resting stage on 1 July, when an apterous form without the paddle-like appendages was found. Edmonson (1959) reported this form to be Polyarthra developed from a resting egg. It is so unique that taxonomists had originally separated it into a separate genus, Anarthra. Many resting eggs and the above described apterous forms were found in the April samples. These resting eggs of Polyarthra also show a thicker wall and numerous shell spines.

Resting eggs from other rotifers in Boulder Basin have not been found.

Daphnia resting stages are commonly called ehippia, named so for the resemblance to a saddle. After formation, the ehippia separates from the adult with the next molt. These ehippia usually contain one to three resting eggs. Stross (1966) reports both photoperiod and water temperature to be controlling factors for the development and release of diapause in Daphnia. Daphnia ehippia were found in the May 1975 and the February through April 1976 samples.

Bosmina did not show a resting stage in our samples, but diapause may play an important role in its life cycle. Bosmina and Daphnia populations occur at different times of the year in Boulder Basin. The fact that Daphnia goes into a dormant state may allow the Bosmina population to become abundant. Hammer and Sawchyn (1971) conclude that

this type of reproductive segregation permits the use and exploitation of an environment by organisms that could not coexist if their reproductive cycles were the same.

Reproductive segregation due to diapause also occurs in the copepods. Cyclops bicuspidatus thomasi and Mesocyclops edax occur at different times of the year, similar to the cladocerans. C. b. thomasi goes into a cyst stage in its fourth instar or copepodid. Cysts of this species were first recorded by Birge and Juday in 1908. Cole (1953) found these cysts in anoxic waters rich in hydrogen sulfide and showed them to be insensitive to various respiratory poisons (NaCN, NaN₃, and idoacetic acid).

It is not known if Mesocyclops edax has an encysted stage, but Smyly (1962) reports cysts of the fifth instar of Mesocyclops leuckarti. Coker (1943) reports these two species to be closely related, with M. edax being the most common North American representative of the genus and M. leuckerti the generic representative for European waters.

Cole (1961) concluded that the resting stage of Cyclops bicuspidatus thomasi may make it possible for it to coexist in the same lakes with Mesocyclops edax. They commonly occur together in North America, with M. edax usually abundant in the summer plankton while C. b. thomasi cysts are abundant in the bottom sediments.

Resting eggs have been found for a few species of Diaptomus, but no resting cysts have been reported (Cooley, 1971). Diaptomus clavipes and Diaptomus siciloides were first reported from Lake Mead by Mildred S. Wilson in 1955 (Cole, 1961). This association is known from other lakes in this region. Cole (1961) reports finding these two species together in large impoundments of the Salt and Gila Rivers of Arizona. He also reports them from a small, newly impounded stock tank in that state. Resting eggs have not been found for either species of Diaptomus from Lake Mead.

Metalimnetic Oxygen Depletion
In Lake Mead

by

John Baker and Thomas Burke

INTRODUCTION

Metalimnetic waters of Lake Mead show a reduction in dissolved oxygen resulting in a negative heterograde oxygen profile. This condition in Lake Mead was first reported by Hoffman et al. (1967). A negative heterograde oxygen profile was evident in 1944 (unpublished data, Bureau of Reclamation) and low metalimnetic oxygen levels probably have occurred since the formation of the lake. Our data, collected as part of the Lake Mead Monitoring Program, has shown that 1) metalimnetic oxygen depletion has been a regular occurrence since 1972, 2) oxygen depletion is always associated with the thermocline, 3) depletion begins in May and continues through September, and 4) hypolimnetic oxygen levels remain high with only minimal oxygen loss during summer stratification.

Mid-water oxygen minimums have been related to three possible causes (Shapiro, 1960): 1) horizontal movement of low oxygenated water due to sediment uptake from a mid-water shelf, 2) density current flows low in oxygen, and 3) oxygen consumption in situ due to biological respiration. The first of these probably does not occur in Lake Mead. There is not an obvious shelf present and the oxygen minimum always develops at the same depth independent of the lake elevation. Density currents have been reported for Lake Mead (Smith et al.

1960). These currents were usually found in the hypolimnion and not in the metalimnion where the oxygen minimum occurs. Hypolimnetic density currents in Lake Mead, if high in dissolved oxygen, may increase the oxygen levels in the hypolimnion, but do not directly cause the mid-water oxygen minimum as Ellis (1940) concluded for Elephant Butte Reservoir, New Mexico. Mid-water oxygen minimums due to biological respiration have been attributed to seston layering, bacteria, and zooplankton. The vertical distribution of bacteria were examined in 1974. Bacterial counts were low and could not account for the oxygen depletion in the metalimnion. A preliminary investigation of the zooplankton in 1974 revealed high concentrations in the metalimnion. Phytoplankton respiration was also suspected as a possible cause. The vertical distribution of both zooplankton and phytoplankton was examined to determine if their respiration could account for the metalimnetic oxygen depletion.

METHODS

The methods for the collection and counting of zooplankton and phytoplankton have been given in previous sections of this report.

It was necessary to determine both the amount of oxygen visibly lost and the amount of oxygen being transported into the metalimnion through eddy currents to obtain the total

amount of oxygen lost from this area. The amount of oxygen being transported into the metalimnion through eddy diffusivity was computed from the following transport equation (Ruttner, 1953):

$$Q_{at} = \frac{D_e(C_1 - C_2)}{L}$$

Where, Q_{at} = the quantity of oxygen (Q) transported across a given area (a) of a horizontal plant (12.5 meters) in some period of time (t).

D_e = the eddy diffusivity or mixing rate.

$C_1 - C_2$ = the difference in oxygen concentrations across this plane.

L = the distance between C_1 and C_2 .

(A practical application of this method is given in Verduin, 1960).

Eddy diffusivity in cm^2/sec was determined from the above transport equation using temperature data, where Q_{at} was the amount of heat transported across this plane. The observed or 'visible' oxygen lost was computed as follows:

$$\text{Visible } O_2 \text{ lost} = \frac{\text{Mean loss of oxygen for some period}}{\text{Number of days in the period}}$$

The quantity of oxygen 'visibly' lost plus the quantity of oxygen transported into the metalimnion equaled the total amount of oxygen to be accounted for.

Calculations for the oxygen consumption of phytoplankton and zooplankton were computed for the metalimnetic populations

Table 17. Calculated respiration rates for limnetic zooplankton in Lake Mead at 20°C.

<u>ORGANISM</u>	<u>ug O₂/day</u>
<u>Copepod Nauplii</u>	0.25
<u>Copepod Copepodites</u> (instars I - IV)	0.80
<u>Cyclops bicuspidatus thomasi</u> (C-V & Ad.)	1.25
<u>Mesocyclops edax</u> (C-V & Ad.)	1.35
<u>Diaptomus siciloides</u> (C-V & Ad.)	1.50
<u>Diaptomus clavipes</u> (C-V & Ad.)	4.50
<u>Daphnia spp.</u>	4.10
<u>Bosmina longirostris</u>	0.86

(10-30 meters) based on reported respiration rates for these organisms. Daily respiration for the major components of the zooplankton community (Table 17) were determined from Comita (1968), Baudouin and Ravera (1972), Shapiro (1960), Bishop (1968), Vollenweider and Ravera (1958), Richman (1958), Scherbakoff (1935), Marshall and Orr (1955), Schindler and Noven (1971), and Kibby (1971). Phytoplankton respiration was determined for estimated cell volumes. A respiration rate of 74 ug/ul of algae/day, calculated from data given in Verduin (1960), was used for phytoplankton respiration.

RESULTS and DISCUSSION

The vertical distribution of zooplankton has been discussed in the previous section of this report. High concentrations of copepods and cladocerans were always found in the zone of oxygen depletion as shown in Figure 12 and total numbers always declined below 30 meters. In general, peak zooplankton concentrations remained within the metalimnion over a 24-hour period. This occurred for all species and their juvenile stages, except for Mesocyclops edax, which nocturnally migrated to the surface waters during July and August. Respiration rates were determined from the evening samples (sunset) which represented resident populations in the metalimnion.

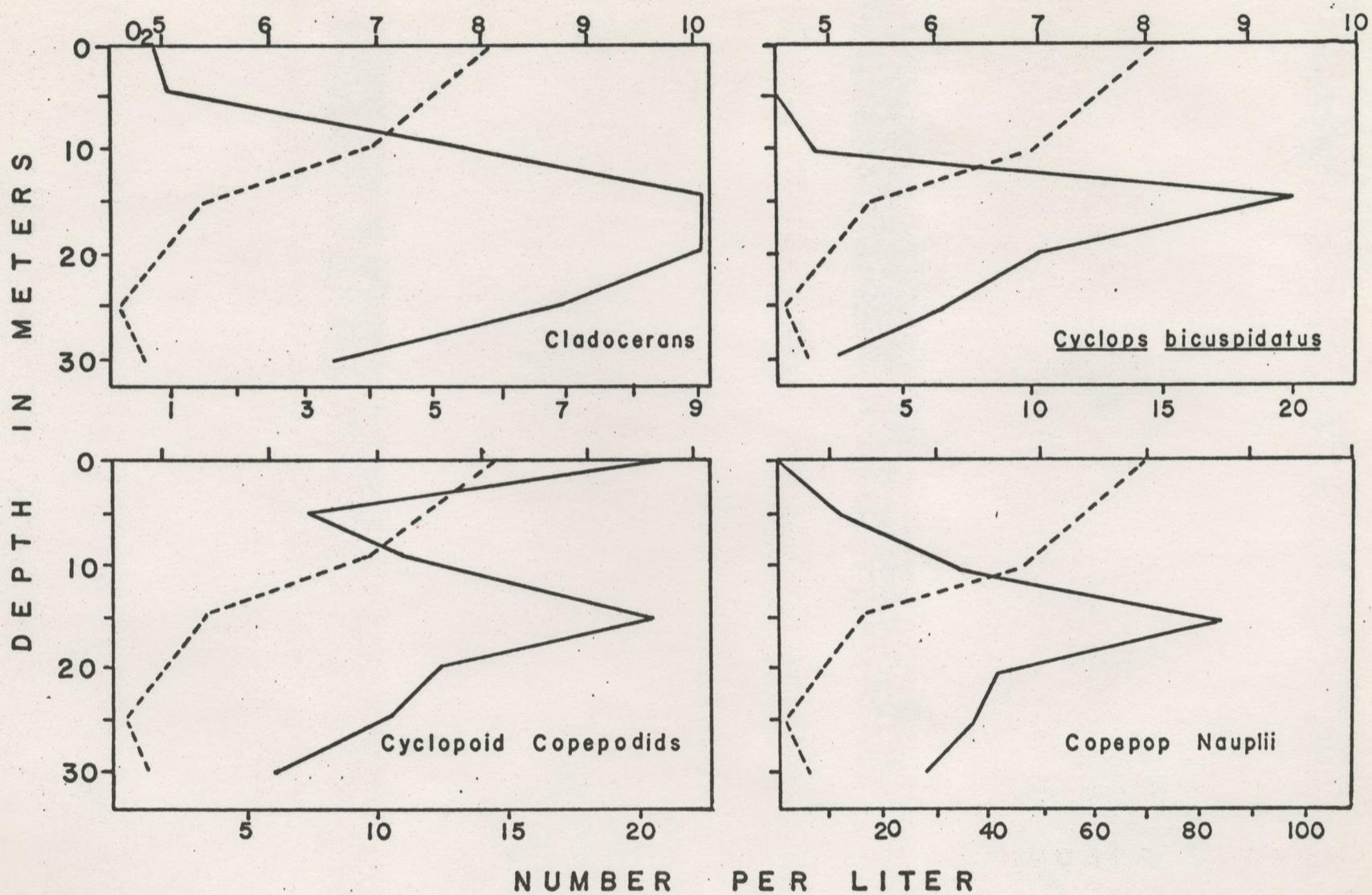


Figure 12. Vertical distribution of zooplankton and dissolved oxygen at Station 5, July 1975. Zooplankton (—), Dissolved oxygen (----)

Phytoplankton numbers were highest in the upper ten meters and declined with depth. There was never an evident seston layer as indicated by phytoplankton counts. Mean phytoplankton volumes between 10 and 30 meters on 16 June, 18 July, 20 August, and 28 August were 0.81, 0.89, 1.33, and 1.03 ml/m³, respectively.

Oxygen transport (Q_{at}) and oxygen 'visibly' lost in the metalimnion during summer stratification were calculated from the data presented in Table 18. Estimated zooplankton and phytoplankton respiration accounted for 57 to 94% of the total amount of oxygen lost during this period (Table 19). Overall, zooplankton accounted for approximately 31% and phytoplankton accounted for approximately 43% of the oxygen lost.

Shapiro (1960) reported that copepods alone accounted for 23 to 110% of the oxygen lost in the metalimnion of Lake Washington and concluded that copepods were responsible for the metalimnetic oxygen minimum in that lake. We are in general agreement with Shapiro's findings, except for his estimated percentage of oxygen consumption by the copepods. Shapiro considered mixing to be minimal and did not determine oxygen transport into the metalimnion. In Lake Mead, mixing results in a substantial amount of oxygen being transported

Table 18. Temperature (°C) and dissolved oxygen (mg/l) at Station 5
17 June through 17 September 1975.

Depth(m)	17 June		2 July		18 July		1 Aug		28 Aug		17 Sept	
	Temp	O ₂	Temp	O ₂	Temp	O ₂	Temp	O ₂	Temp	O ₂	Temp	O ₂
0	22.5	10.1	22.5	9.9	24.5	7.4	26.0	8.1	25.5	8.3	26.0	8.9
5	21.5	10.4	22.5	9.9	24.5	7.4	26.0	7.4	25.5	8.0	26.0	8.6
10	20.0	9.4	21.5	9.4	23.5	7.2	25.0	6.8	25.0	6.0	25.0	4.5
15	17.0	7.7	20.0	8.1	20.5	5.9	21.5	5.3	23.0	4.0	24.5	3.6
20	15.0	7.4	16.5	7.2	17.5	5.1	18.5	4.8	20.5	4.1	21.5	2.2
25	14.0	7.4	15.0	6.9	15.5	5.0	16.5	4.5	18.0	3.8	19.5	3.5
30	13.0	7.9	13.5	7.0	14.5	5.3	15.5	4.7	16.0	4.2	17.5	3.5
35	12.5	8.0	13.0	7.2	13.5	5.5	14.0	5.3	15.0	4.6	15.5	4.9
40	12.0	8.0	12.5	7.5	13.0	6.0	13.0	5.7	14.5	5.5	14.5	5.5
45	11.5	8.0	12.0	7.7	12.0	6.3	12.5	5.9	13.5	5.9	13.5	5.9
50	11.5	7.9	11.5	7.7	12.0	6.3	12.0	6.2	13.0	6.5	13.0	6.5
55	11.5	7.9	11.5	7.7	11.5	5.8	11.5	6.4	12.5	6.6	12.5	6.6
60	11.0	7.9	11.0	7.7	11.5	--	11.5	6.3	12.5	6.2	12.5	6.5

Table 19. Total amount of oxygen lost (oxygen transported in plus oxygen 'visibly' lost) and respiration rates for zooplankton and phytoplankton within the metalimnion. All units are in micrograms of oxygen per liter of water (ug/L). Numbers in parenthesis equal the percentage of oxygen possibly consumed due to zooplankton and phytoplankton respiration.

	17 June to 2 July	to 18 July	to 1 Aug.	to 28 Aug.	to 17 Sept.
Oxygen Transported (Q_{at})	118	70	64	110	129
Oxygen 'Visibly' Lost	23	126	39	32	55
Total Oxygen Lost	<u>141</u>	<u>196</u>	<u>103</u>	<u>142</u>	<u>184</u>
Estimated Zooplankton Respiration	60 (42%)	51 (26%)	47 (46%)	52 (36%)	30 (16%)
Estimated Phyto- plankton Respiration	59 (41%)	66 (33%)		83 (58%)	75 (41%)

into the metalimnion. For some periods, zooplankton respiration accounted for over 200% of the oxygen 'visibly' lost. Adding the amount of oxygen transported into the metalimnion to the computations reduced this to a realistic value of 31%.

Zooplankton respiration rates were conservative estimates and these rates may be higher than those reported. This may have little effect on the results as Verduin (personal communication) feels our mixing rates are also conservative because they were computed for bi-weekly periods. Mixing rates or eddy diffusivity computed on a daily basis are usually higher than those computed over an extended time interval.

Other organisms were found within the metalimnion, and their respiration could possibly account for the remaining amount of total oxygen lost. Respiration rates for the rotifer were not determined because peak concentrations generally were found above ten meters, but individuals did occur within the metalimnion in low concentrations (Table 20). Echograms showed large concentrations of shad within the metalimnion during the early morning (Fig. 13). The shad were dispersed throughout the epilimnion in the afternoon and were concentrated in a narrow band at the surface nocturnally. While these organisms were present

Table 20. Rotifers (#/l) at Station 5, Boulder Basin
July 1, 1975.

<u>1530 hours:</u>	<u>Depth (meters)</u>						
	0	5	10	15	20	30	40
Rotifers							
Asplanchna	0.5	3.5	11.0	0.5			
Polyarthra	2.0	8.0	3.0	0.5	0.5		
Collotheca	21.0	21.0	4.0	1.0	0.5	0.5	0.5
Keratella cochlearis	0.5	3.5	2.0	0.5	--	0.5	0.5
Keratella quadrata	--	--	1.0	0.5	0.5		
Syncheata	11.0	10.0	5.0	1.0	0.5		

<u>2030 hours:</u>	<u>Depth (meters)</u>						
	0	5	10	15	20	30	40
Rotifers							
Asplanchna	1.0	3.5	14.0	1.0			
Polyarthra	2.0	10.0	3.0	4.0	0.5		
Collotheca	15.0	19.0	8.0	5.0	1.5		
Keratella cochlearis	0.5	3.0	4.0	--	--	0.5	0.5
Keratella quadrata	--	--	2.0	0.5	0.5	--	0.5
Syncheata	2.5	18.0	0.5	--	0.5		

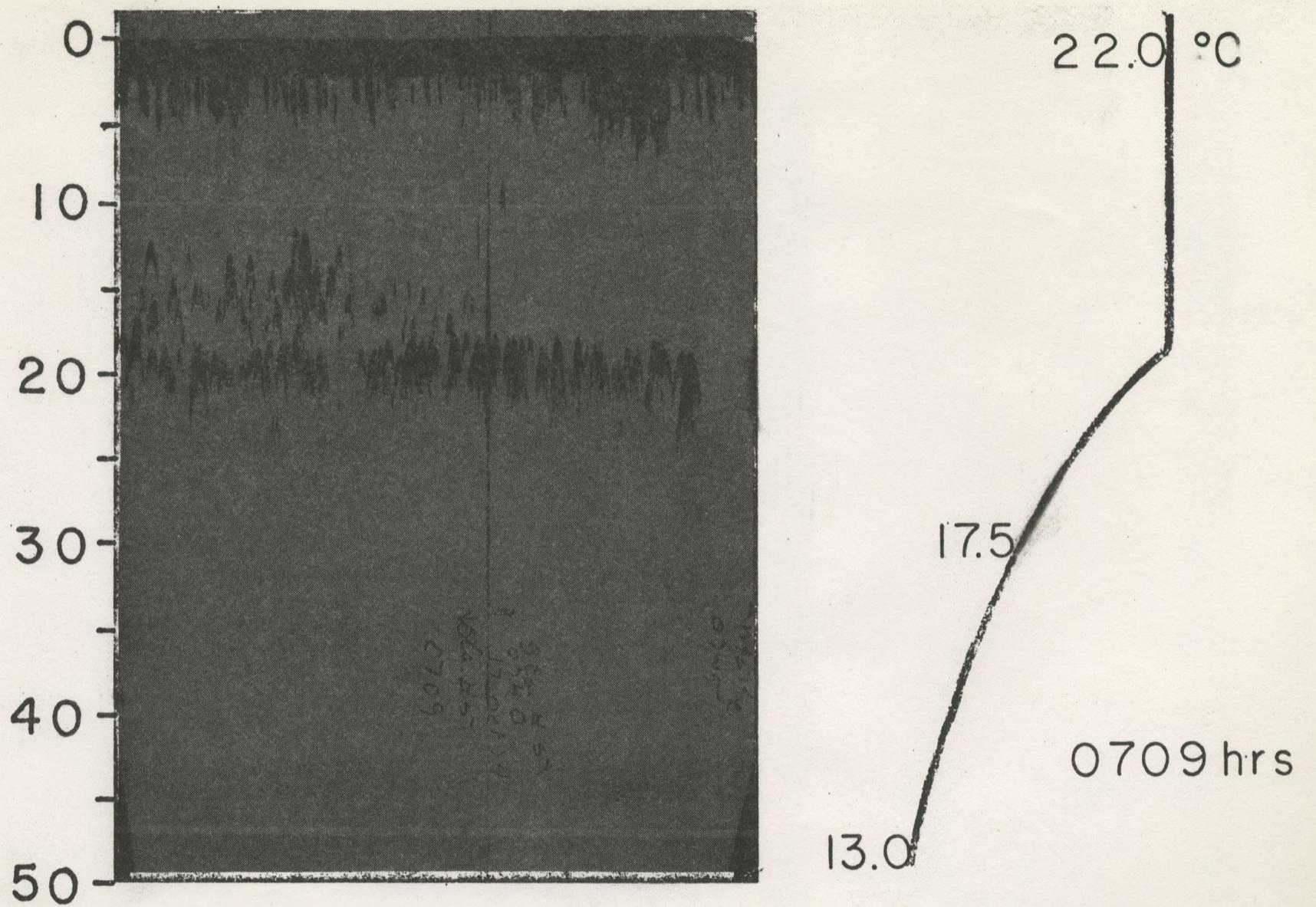


Figure 13. Ecogram of shad distribution and temperature profile at Station 5, October 1975.

within the metalimnion either in low numbers or for short periods of time, their respiration would result in further loss of oxygen from this area. The results indicated that biological respiration could account for the metalimnetic oxygen depletion and that phytoplankton and zooplankton respiration are the primary causative agents.

Phytoplankton and zooplankton communities in Lake Mead are not strikingly different from other eutrophic lakes. The primary factor resulting in the negative heterograde oxygen profile is the deep hypolimnion and minimal oxygen uptake of the mud-water interface. Oxygen depletion in the metalimnion affects only the upper portion of the hypolimnion because of reduced turbulence during summer stratification. At Station 3, which has a maximum depth of 45 meters, and at other shallow areas, a clinograde oxygen curve does develop. If Lake Mead was more shallow, a typical clinograde oxygen profile would probably occur.

Coliform Bacteria in Las Vegas Wash
and Las Vegas Bay

by

Sam Egdorf

INTRODUCTION

The coliform group of bacteria have been generally accepted as indicators of sanitary quality, and as standards for general use. These indicator organisms are assumed to indicate the degree of fecal pollution in water.

Standards for coliforms in potable and recreational water have been established by various agencies. Counts of these bacteria may be interpreted to indicate compliance or non-compliance with these standards.

Enforcement of coliform water quality standards depends on the validity of the methods employed for coliform enumeration. This investigation incorporated organism identification (Cowan, 1974) and media comparisons to insure reliable estimates of coliform concentration.

METHODS

Coliform enumeration - Water samples were collected at Stations 1-3 in a 3 liter Van Dorn bottle previously sanitized with acetone-alcohol. Samples were transferred to a screwcap, glass bottle, placed on ice, and delivered to the laboratory for enumeration of fecal and total coliforms by the membrane filter procedure (American Public Health Association, et al., 1971).

Coliform survival in sediments - Four gallons of water was collected, at a point just above the convergence of Las Vegas Wash with Las Vegas Bay, in previously sanitized glass collection bottles, placed on ice, and delivered to the laboratory. All four samples were combined and one liter was placed in each of four Imhoff funnels. The sediments were allowed to settle and incubated at 25°C. A fifth aliquot was shaken and evaluated for total coliforms by the membrane filter procedure. Two days after collection two Imhoff funnels were evaluated for coliform concentrations in supernatant and sediment by the membrane filter procedure. Fifteen days after collection the remaining Imhoff funnel was enumerated for total coliforms in both sediment and supernatant by the membrane filter procedure.

RESULTS and DISCUSSION

Both fecal and total coliform populations were low in April. An increase in their incidence was noticed in May and continued through June (Table 21).

During July, a flash flood damaged the City of Las Vegas Sanitation Plant. The flood resulted in greatly increased coliform concentrations in Las Vegas Wash (LVW) and Las Vegas Bay (LVB). However, during this period higher concentrations of enteric bacteria were found to be Erwinea

Table 21. Fecal and total coliform bacteria (#/100ml) in Las Vegas Wash and Las Vegas Bay.

Station	Depth	<u>28 April</u>		<u>16 May</u>		<u>28 May</u>		<u>18 June</u>		<u>28 June</u>		<u>22 July</u>	
		Fecal	Total	Fecal	Total	Fecal	Total	Fecal	Total	Fecal	Total	Fecal	Total
1	0	60	80	800	230*	300	2100	100	270	90	1600	1500	2190
2	0	0	0	30	2*	0	0	20	7*	1	3	0	4
	10	40	30*	0	100	10	30	11	270	10	230	8	280
3	0	0	0	0	1	0	1	0	0	5	2*		
	10	0	1	0	2	0	0	0	0	0	0		
	20	0	0	0	0	0	0	0	0	1	12		
	30	0	0	0	1	0	1	0	2	0	0		
	40	0	0	0	0	0	0	0	1	1	3		
		<u>31 July</u>		<u>19 Aug</u>		<u>28 Aug</u>		<u>17 Sept</u>		<u>16 Oct</u>		<u>24 Nov</u>	
1	0	1700	1900	700	2000	10	2430	0	0	50	900	180	1000
2	0	0	0	7	2*	30	134	0	0	0	12	0	7
	10	70	140	7	25	10	103	0	300	0	30	2	41
3	0	0	0	0	0	0	150	0	0	0	0	0	3
	10	0	0	0	8	0	1300	0	0	0	0	0	23
	20	0	4	0	4	0	1400	0	0	0	3	12	30
	30	0	100	0	50			0	20	0	0	0	5
	40	20	650	0	300			0	8	0	4	0	76
		<u>22 Dec</u>		<u>30 Jan</u>		<u>19 Feb</u>		<u>30 Mar</u>					
1	0	10	120	0	0	0	140	0	70				
2	0	0	10	10	700	0	0						
	10	0	9	1	800	0	0						
3	0	0	2	0	120	0	0						
	10	0	0	0	240	0	0						
	20	0	0	0	80	0	0						
	30	0	3	1	200	0	0						
	40	0	2	1	180	0	0						

herbicola and Klebsiella pneumoniae. E. herbicola is generally associated with plant galls, while K. pneumoniae is found in association with root systems along with fecal material (Cowan, 1974). Because such a large volume of water came down the wash during this period, and because the largest number of enteric organisms found may have been associated with plants, the source of these bacteria may have been from the marsh system and not from the sanitation plant. However, this observation is conjecture because sampling was not continuous during the breakdown of the sanitation plant.

Prior to the July flash flood, lake drawdown caused erosion and redistribution of sediments. It was felt that sediment re-distribution may re-suspend sediment-bound organisms of coliforms entering Las Vegas Bay via Las Vegas Wash. However, the persistent effects from the flood curtailed continued examination.

In September, coliform counts were low with the exception of Station 2 at 10 meters. Relatively high counts were obtained in October and November. Counts were low in December but an increase was noted in January 1976. However, February and March 1976 coliform counts were low and comparable with April 1975.

Normally, the fecal to total coliform ratio is less than one, however, FC/TC ratios of greater than one were found at various times and locations during the year (Table 21). Causes for the noted anomalies have not been determined. Technical procedures have been eliminated as a causative factor. Possible inhibitory and toxic effects of plating media (m-Endo m-FC) have not been eliminated conclusively and, therefore, are still implicated.

Data from the current study and from the 1974-75 monitoring program indicate a significantly greater survival rate for K. pneumoniae than E. scherichia coli on these media. A comparison between m-Endo and m-Endo-LES (Table 22) indicates a significantly greater survival of coliforms plated on Endo-LES. Obviously, further investigation is required to determine which of the recommended coliform media is least detrimental to coliform survival.

Samples collected in LVW from Sunrise Power Station to the convergence of LVW with LVB resulted in relatively low fecal and total coliform counts (Table 23). However, samples were collected between 7:00 and 10:00 a.m. and may not have been during peak flow periods from any of the sanitation plants. Further sampling of LVW should be conducted during peak flow periods and extended over a period of several months to determine the major sources of coliform introduction

Table 22. Media comparison m-Endo, m-Endo-LES and m-FC
(#/100ml).

Depth Meters	Station 1			Station 2		
	m-Endo	m-Endo-LES	m-FC	m-Endo	m-Endo-LES	m-FC
0	2.19×10^3	7.6×10^3	1.5×10^3	3.6	72	0
10				2.08×10^2	3.62×10^2	8.2

Table 23. Total and fecal coliforms in Las Vegas Wash (#/100ml).

<u>Location</u>	<u>Total coliforms</u>	<u>Fecal coliforms</u>
Sunrise Power Station	2	0
Las Vegas Sewage Plant	48	18
Clark County Sewage Plant	37.5	20
Pabco Road	84	24
Above Gravel Pit	0	0
Gravel Pit	0	0
Las Vegas Bay Estuary	45	18

to LVW. Sample points should also include the contribution of marsh areas as well as other streams flowing into LVW.

The incidence of Salmonella and Shigella in LVW and the inner bay of LVB was determined during the July flash flood. Although Salmonella and Shigella cannot be determined quantitatively, their presence or absence can be determined. Salmonella was detected at Stations 1 and 2.

The survival of sediment bound coliforms was significant (Table 24). Although results from the laboratory study performed can not be applied directly to a lake situation, certain empirical conclusions may be reached. 1) Coliforms survive significantly longer in sediments. 2) The initial coliform population will increase in size in nutrient rich sediments. 3) Resuspended bottom sediments may be a cause for increased coliform densities in LVB.

Table 24. Survival of sediment bound coliforms from Las Vegas Wash (#/100ml).

Days	Supernatant	Sediment
0	520	
2	140	800
15	9.2	6.72×10^3

Distribution of Enteric Bacteria in Las Vegas Bay

by

Sam Egdorf

INTRODUCTION

The utility of oxidase positive bacteria for tracing water distribution patterns has been established (Tew et al., 1976). Although these bacteria are a large component of the bacterial population, there is another component of the population composed of oxidase negative bacteria, among which are included those Enterobacteriaceae of special medical significance (Salmonella, Shigella, Klebsiella, Yersinia).

Also, media used for the isolation of oxidase negative bacteria are inhibitory and usually result in low population estimates, while in fact these bacteria may be present in numbers much higher than one would expect.

For these reasons it was decided to look at the oxidase negative bacteria as a means of determining water distribution patterns. Data concerning their deposition in the lake could perhaps be obtained simultaneously.

Previous students of water distribution patterns did not use a confirmed method to substantiate their results (Hoganson and Elliot, 1972; Losane et al., 1967; McFeters et al., 1974; Storey et al., 1974). However a study conducted in 1899 (Jordan, 1900) (to determine the extent

of natural purification of fecal bacteria by lakes and streams) did utilize chlorine and fluorescein dye as a confirming technique in conjunction with bacterial sampling. Although the supporting methods did not correlate directly with bacterial concentrations, they did indicate water distribution patterns from a specific sewage source.

Flourescent dyes, because of their relatively high fluorometric detectability, should provide an excellent precursor for bacterial sample collections. An instantaneous or "slug" injection into a moving body of water should produce a well defined dye peak that could be easily monitored by fluorometric methods as the dye moves away from the point of injection (St. John, 1973; Storey et al., 1974). The dye peak would indicate water velocity, dilution rate and provide a signal to insure sampling of a specific bacterial population as it progresses away from an initial sampling point.

Inhibitory effects of differential media on oxidase negative bacteria have been discussed extensively in the literature (Bascomb et al., 1973; Dufour and Cabell, 1975; Hartman et al., 1975; Hoganson and Elliot, 1972; McCoy and Seidler, 1973; Ray and Speck, 1973). The results of these investigations indicate that utilization of non-inhibitory media may be instrumental in isolation of far greater numbers

of oxidase negative bacteria than could be achieved with differential media. Obviously the use of noninhibitory and non-differential media would make characterization of the enteric genera more difficult, but replica plating methods (Lederberg and Lederberg, 1952) and biochemical characterization on initial isolation plates (Dufour et al., 1975) might facilitate characterization greatly. Difficulties in enteric identification created by the use of non-inhibitory media would be far outweighed by the increased harvest of these bacteria.

The distance to which bacterial tracing can be accomplished with confidence will depend on the initial tracer population (ITP) amplitude and duration. Past data has indicated that ITP may last from a few minutes to several days. Also the concentration of enteric bacteria may vary from 0/100 ml LVW sample to as high as 10^4 /100 ml. Previous studies (Tew et al., 1976) have demonstrated that individual tracer bacteria concentrations of greater than 10^5 /100 ml over a finite period of time are required for extensive tracing of water distribution patterns in Las Vegas Bay, for one reason to overcome the noise created by a given count of indigenous lake bacteria, for another, to obviate sedimentation and biological decay with specific reference to enteric bacteria. Other precepts were that tracer bacteria should not be indigenous to the lake and should appear in LVW periodically. Regarding the latter there is also the possibility that resuspension of sediment bound organisms may

perturb ratios of organisms found to some extent (Cook et al., 1974; Grimes, 1975; Hendricks and Morrison, 1967; Hendricks, 1971, 1972).

Previous studies conducted in Lake Mead indicate that there may be residual population of these organisms present in the lake at all times. Assuming this is true, the entering enteric population from Las Vegas Wash would have to be in concentrations high enough to overcome the noise created by the residual enteric populations for any extensive tracing in Las Vegas Bay.

Enumeration of the individual genera and species in the population may serve as a second confirmatory means to insure that the same incident population is being sampled at all times. Also they are an excellent way of studying the concept that tracers need not be intermittent to be used, if two or more of them vary in relative numbers consistently over a given period of time ("Component Ratio Concept"). For example, if organism A is usually twice as numerous as B, then suddenly, for two weeks or so becomes half as numerous as B then the change in ratio constitutes a traceable situation.

MATERIALS AND METHODS

Sampling locations were selected from midstream of Las Vegas Wash to a point approximately 1400 meters below the

convergence of the wash with Las Vegas Bay. The sampling locations are illustrated in Figure 14.

Dye injection and detection. One gallon (3.785 l) of Rhodamine WT which was determined to be the same density as the water in the wash (1.025 g/ml) was placed in a glass container, suspended in midstream of Las Vegas Wash at Sampling Point 1 and allowed to equilibriate for 30 minutes. The container was then broken and an instantaneous dye injection was achieved.

Because of the shallow water at Sampling Point A (less than 30 cm) the boat containing the fluorometric detection equipment could not be utilized for continuous sampling of the dye peak. Grab samples were collected at five minute intervals and returned to the boat for analysis. Samples were collected until 15 minutes after the dye peak passed the sampling point. Detection of the dye was accomplished with a Turner Model #111 Fluorometer, the output of which was recorded on a stripchart recorder. At Sampling Points G and 2 the complete dye envelope was monitored and recorded. This was accomplished by suspending a hose into the water column at density current depth, a depth established by conductivity readings and pilot dye studies. The hose was connected to a D.C. self-priming pump, the output of which was connected by 3/8 inch tygon tubing to the flowthru cell of the fluorometer.

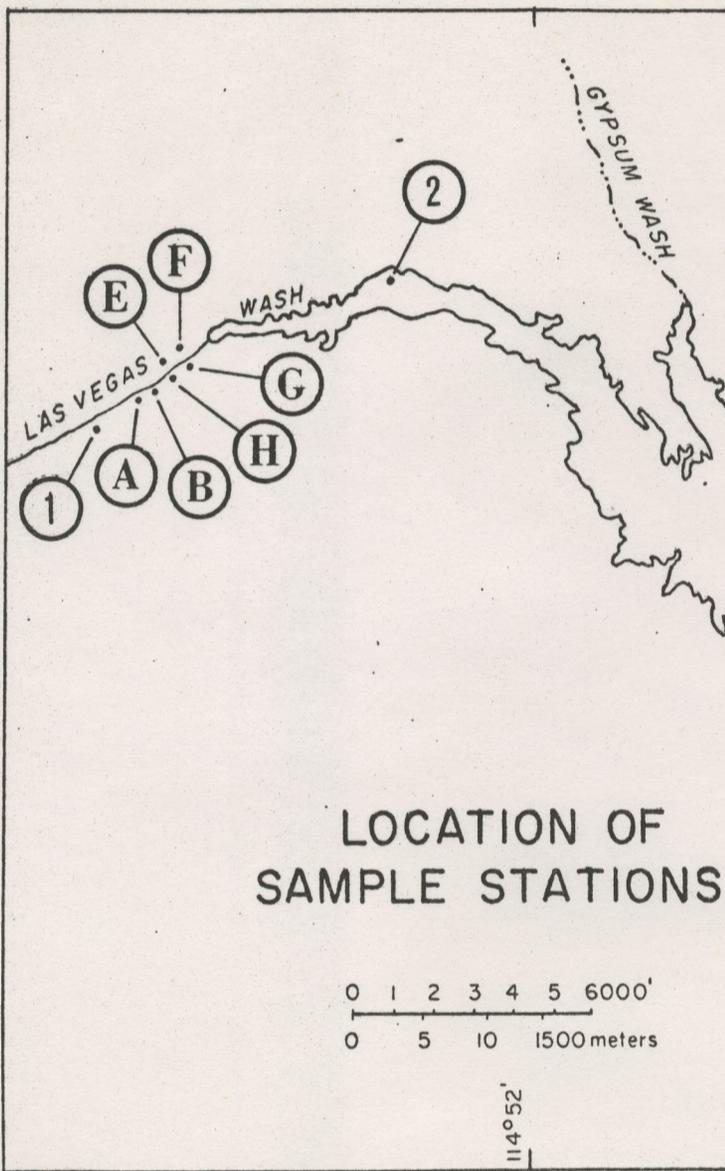
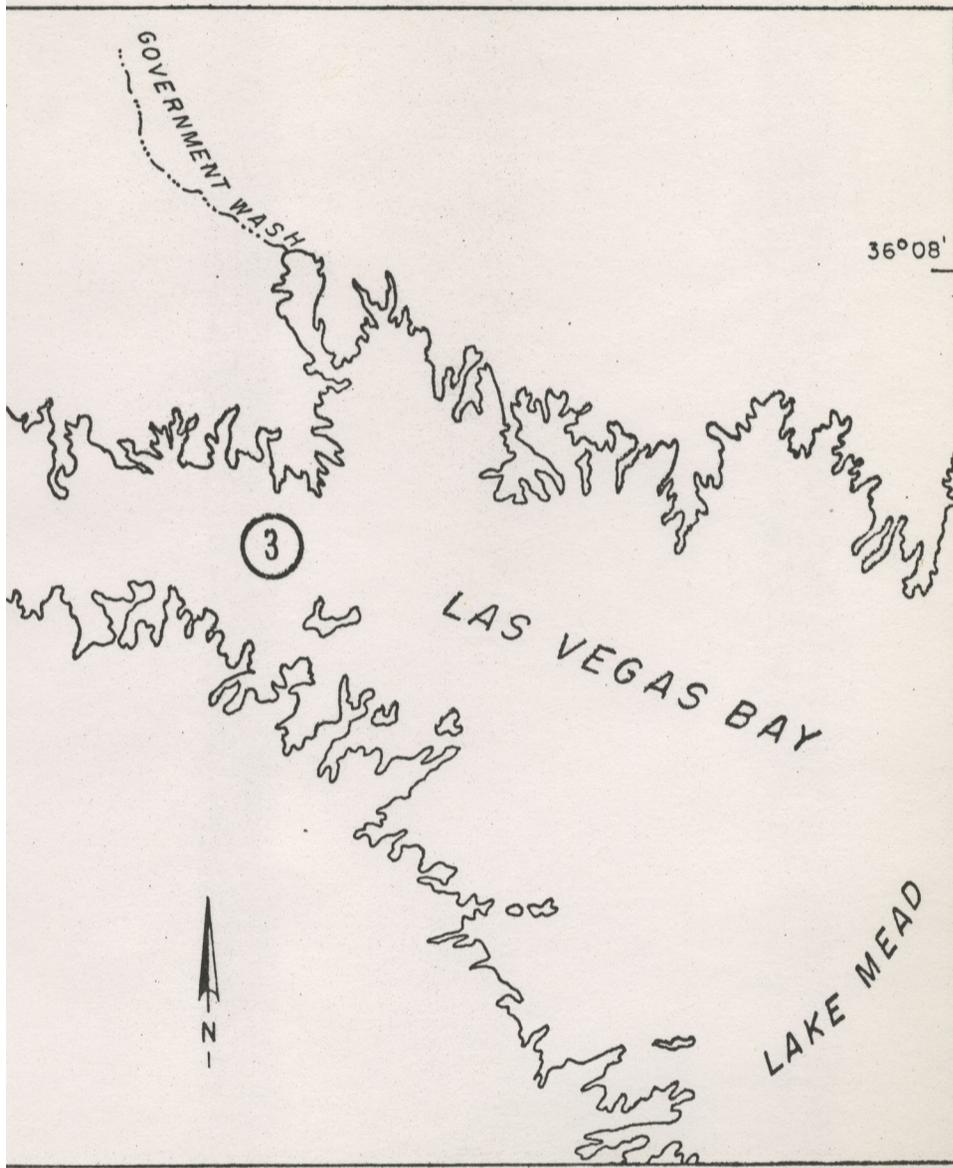


Figure 14. Sample Point locations



in Las Vegas Wash and Las Vegas Bay.

Bacterial sample collection. Bacterial samples were collected at the dye injection point 15 minutes after dye injection and 15 minutes after dye peak arrival at all other sampling points. The samples were collected in this manner to insure against the possible toxic effects of the dye on the organisms. Also this method theoretically would insure continuous sampling of a discrete bacterial population at each sampling point.

Samples collected at Sampling Points 1 and A were collected with a three liter Van Dorn bottle previously sanitized with acetone-alcohol. Because of the shallowness of the water at 1 and A and because of the rapid flow only one subsurface grab sample was taken. Samples collected at Sampling Points G and 2 were collected with a previously sanitized three liter Van Dorn bottle. Samples were collected at the surface and 3.5 meters at G, and at the surface and 10.5 meters at 2 (samples collected at 3.5 meters and 10.5 meters had been previously determined as the vertical center on the density current at these sampling points). Immediately upon collection the bacterial samples were transferred to glass screwcap bottles, placed on ice and transported to the laboratory for analysis. Note that the time from collection to initial isolation of the bacterial samples did not exceed three hours.

Bacterial isolation. Initial isolation of the samples was by the membrane filter method (American Public Health Association et al., 1971). Sample volumes filtered were 100 ml thru 0.0001 ml in dilutions of ten. All dilution volumes less than 1 ml were obtained by diluting 10:1 in 9 ml peptone water dilution blanks. The samples were filtered in two sets of five replicates for each dilution.

One set of filtered samples were placed on pads saturated with m-Endo media and then incubated at 35°C for 24 hours. The remaining set of filtered samples were placed on pads saturated with nutrient broth (NB) and incubated and were incubated at 27°C for 48 hours (Dufour and Cabel, 1975). After 24 hours the membranes incubated on m-Endo were enumerated for typical coliform colonies of dilutions containing 7 to 70 colonies per plate. Typical metallic sheened colonies were isolated onto EMB agar plates for tentative identification and culture purification. All colonies demonstrating typical characteristics were then identified to species. The second set of plates were enumerated for total bacterial growth at the end of incubation, replica plated onto violet red bile agar (VRB) (Lederberg and Lederberg, 1952) and incubated at 35°C for an additional 24 hours. After incubation the plates were enumerated for

typical colonies. All typical colonies were isolated onto EMB agar for colonial purification. Both typical and atypical colonies were then identified to species.

m-Endo media was included in this experiment as a standard referenced method. Also note that identification of isolates from each of the media would result in information concerning its specificity, or inhibitory action, for certain tribes or specific genera of the enteric group.

Identification of isolates. All isolates were first evaluated for their oxidase reaction. Colonies growing on filters incubated on sugar free NB were subjected to the oxidase test of Daubner and Mayer (Dufour and Cabell, 1975), with the test reagent being applied directly to absorbant pads and placing the filter on the pad. All colonies demonstrating oxidase negative reactions were enumerated and their corresponding position on the replica plate was recorded. Isolates from m-Endo were transferred to duplicate NB slants and oxidase reactions were performed in the slant only. Pure cultures of isolates from VRB were also transferred to NB slants and oxidase reactions were performed in the slant only. Pure cultures of isolates from VRB were also transferred to NB slants and the oxidase test was reconfirmed. All isolates were tested for gram

reactions and their ability to ferment dextrose. All isolates exhibiting oxidase negative, gram negative, and dextrose fermentation characteristics were subjected to the multimedia identification scheme.

RESULTS

The dye peak was easily monitored. Its transit time from initial injection to Sampling Point A was 45 minutes. The distance from Sampling Point 1 to Sampling Point A is 1200 meters, thus water velocity from 1 to A was 0.444 m/sec.

From Sampling Point A to G dye transit time was one hour and fifty minutes. The distance from Sampling Point A to G is 400 meters and the water velocity is 6.06×10^{-2} m/sec. The force required for this deceleration, assuming the density of the water is 1.025 g/cc, and calculated by the formula $F = \Delta v / \Delta t \times m$, is 5.8×10^{-3} dynes.

The water column temperature at G is isothermal from surface to bottom. Obviously if the energy of deceleration is not given up as heat, it must be conserved as turbulence resulting in redistribution of bottom sediments.

Transit time of the dye from Sampling Point G to 2 was four hours and five minutes, representing a distance of 1000 meters and a water velocity of 6.06×10^{-2} m/sec. Between

G and 2 the density current encounters a significantly larger volume of water than was encountered prior to reaching Point G, yet in 1000 meters between G and 2 there was no decrease in water velocity. This would indicate that the turbulent water is not mixing as it penetrates further into the lake, but acts as though it is being held in a tight column, a situation not unlike that in a pipe. (An attempt was made to trace the dye peak further into Las Vegas Bay but it was never detected beyond Sampling Point 2).

No attempt was made to determine the dye concentration at Sampling Point A but concentrations at Sampling Points G and 2 were 1.095 ppm and 0.44 ppm respectively. The dye dilution factor indicated at Sampling Point G was $9.13 \times 10^{11}:1$ while at Point 2 the dilution factor was $2.28 \times 10^{12}:1$. The increase of dilution from G to 2 was only a factor of approximately 2:1, which appears to substantiate the theory that the density current is relatively unaffected as it proceeds from G to 2. The leading edge of the dye envelope was much sharper at Point G than at point 2 (Figure 15). The sharp peak at G and the decreased slope of the envelope at 2 indicates decreasing turbulence from G to 2. Note that the dye duration is approximately four minutes shorter at 2 than at G.

Because of the high background fluorescence in the lake, the actual leading and trailing edges could not be

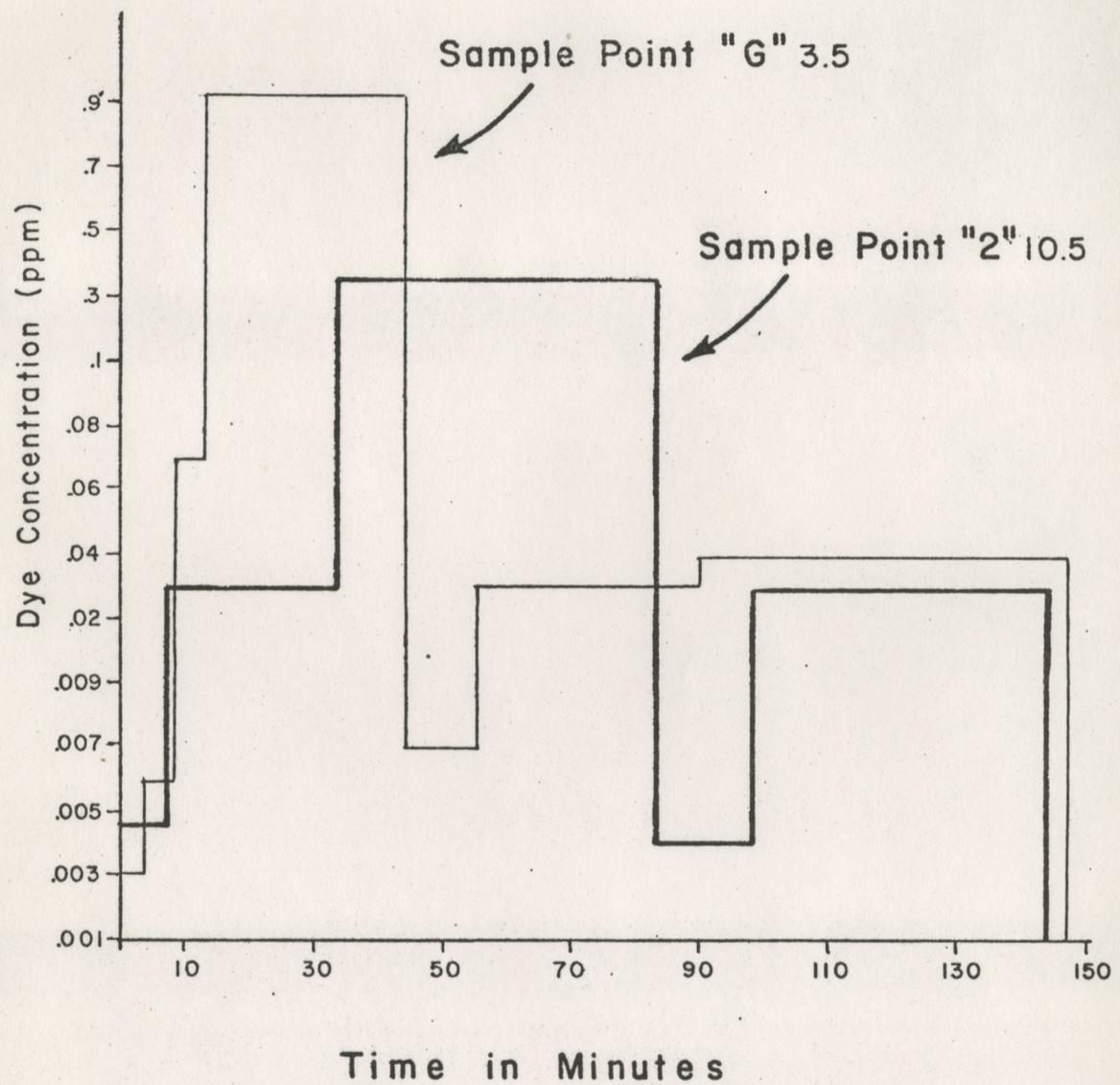


Figure 15. Integrated concentrations of the Rhodamine WT dye envelope detected in the density current at Sample Points G and 2.

determined with a great deal of accuracy. The dye envelope may in fact be significantly longer than indicated fluorometrically. The shape of the dye peaks indicate that there is more turbulent flow at Sampling Point G than at 2.

The apparent dye peak broadening would seem to preclude the coincident sampling of any incident population initially correlated with dye in the stream. In fact, bacterial samples collected at sampling points did not reflect the same dilution pattern as the dye (Table 25). Between Sampling Points A and G there was a decrease in bacterial concentrations (both total bacteria and enteric bacteria) and between Points G and 2 there was an increase in the bacterial populations.

Surface samples collected at Sampling Point G and 2 contained bacterial concentrations of the same order of magnitude, which indicates that there may be a residual population in the lake at all times. Prior to this investigation a bacterial study was conducted between Sampling Point A and G (Table 26) during a period when enteric concentrations were approximately $1.4 \times 10^4/100$ ml (determinations were on the basis of m-Endo isolation media) and during this time the surface concentrations were of the same order of magnitude as when incoming concentrations

Table 25. Counts of total bacteria versus enteric bacteria at Sample Points A, G, and 2.

Sample Point	Depth (Meters)	Total Bacteria Per 100 ml (NB)		Enteric Bacteria Per 100 ml (VRB)		Enteric Bacteria Per 100 ml (m-ENDO)	
		\bar{x}	δ	\bar{x}	δ	\bar{x}	δ
A	0	3.73×10^7	2.82×10^6	1.04×10^6	2.79×10^5	5.6×10^2	5.5×10^1
G	0	9.5×10^4	2.13×10^4	6.6×10^3	4.9×10^3	0	0
G	3.5	6.7×10^5	8.52×10^4	8.6×10^4	1.67×10^4	78	16
2	0	8.8×10^4	5.5×10^3	7.0×10^3	3.3×10^3	3.4	1.14
2	10.5	1.06×10^7	5.45×10^5	5.8×10^5	1.79×10^5	24	11.4

Table 26. Counts of enteric bacteria at Sample Points A and G during a period of high concentration in Las Vegas Wash.

Sample Point	Depth (Meters)	Enteric Bacteria Per 100 ml	
		\bar{x}	σ
A	0	1.12×10^4	1.68×10^3
G	0	22.5	3.6
G	3.7	5.44×10^3	4.3×10^2

of enterics were at $5.6 \times 10^2/100$ ml. There was, however, a change in the ratios of the component species in the population.

Total bacterial concentrations were consistently higher than enteric concentrations isolated on either VRB or m-Endo. Enteric concentrations were consistently higher on replica plated VRB than on m-Endo. There was a slight error in the enumeration of oxidase negative bacteria based on the results of the oxidase reaction on colonies grown on NB media. A significant number of the oxidase negative colonies were actually very weak oxidase positive bacteria. Therefore the differential count of total bacteria versus enteric bacteria was similarly affected.

The comparison between m-Endo and VRB media (to determine the extent to which m-Endo was inhibitory to enterics) was a complete success. Data concerning total numbers of enterics isolated on each of the media confirmed literature conclusions that VRB was less inhibitory. However, data on the specific genera affected to the greatest extent was not determined because of the choice of EMB as a purification medium. Although widely used as a recommended medium for this purpose, EMB turned out to be as inhibitory as m-Endo to most of the isolates from VRB and resulted in a

markedly reduced percentage of identified colonies. This is another example of the unreliability of thoroughly referenced methods. However, subsequent identification procedures using standard multimedia techniques were eminently satisfactory.

DISCUSSION

Optimal tracing of bacterial distributions depends primarily on the detection of a unique population component present over a finite period of time consistent with requirements predicted from the systems analysis.

An intensive search revealed no unique component in the bacterial population. In view of the fact that greater than 500 isolates were identified, this conclusion is eminently justified. The results did indicate that varying ratios of consistently present species might be equally useful.

The results also provided an excellent picture of the enteric bacteria actually present, a result quite significant for public health evaluation.

For example, consider the high numbers of Klebsiella pneumoniae, a known pathogen present in the wash(Figure 16). From a public health point of view it is fortunate that greater than 50 percent of these organisms are sedimented out at the convergence of LVW with Las Vegas Bay. Also it is equally important that large numbers of dysentary

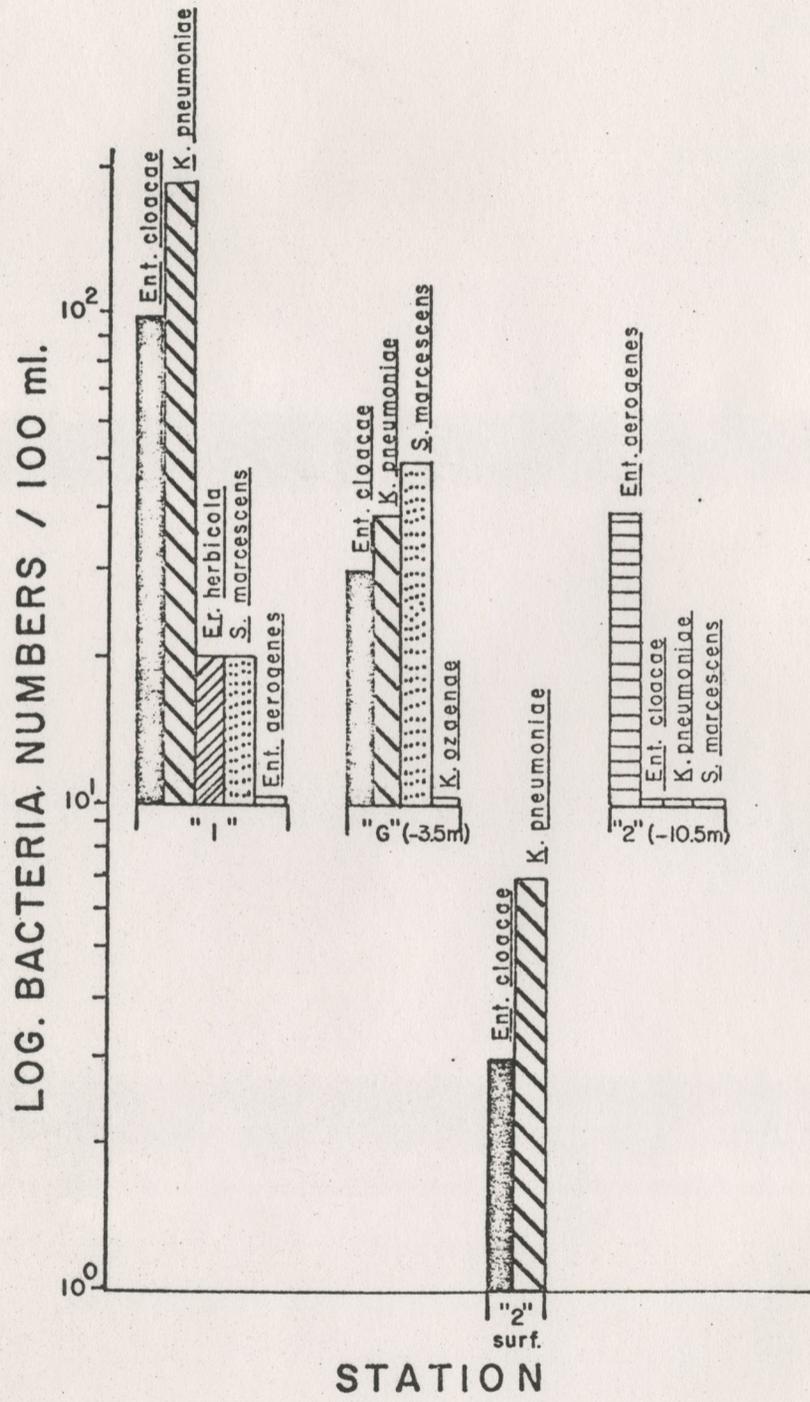


Figure 16. Component enteric tracer bacteria detected in the density current and on the surface.

(Shigella sp.) and enteric fever (Salmonella sp.) bacteria were not found, although they have been demonstrated to be present in numbers far less than an infectious dose (10^6) per unit volume of water. Additional work concerning the fate of sedimented pathogens is strongly advised.

Returning to the basic discussion of water and bacterial distributions, it is felt that differential count of total numbers of LVW enterics versus oxidase positive bacteria, and the proportionate distribution of constituents comprising LVW and residual lake enteric populations provided a valid tracing procedure.

The preceding conclusion was fortified by coincident substantiating experiments conducted with Rhodamine WT. Both the bacteria and the dye 1) went to the same place, 2) were influenced by similar factors, turbulence and sedimentation (biological decay was an additional factor affecting bacteria only), 3) both were reduced in their respective concentrations, however the bacteria were affected to a greater extent than Rhodamine WT. The results however, demonstrate that this unfavorable relative reduction could be more than compensated for by the use of bacteriological procedures of much greater sensitivity.

Inhibitory effects of media. It is abundantly obvious that the inhibitory nature of the standard media totally

counterindicates their further use and also thoroughly justifies the concern of this study with optimized techniques.

The inhibitory effects of m-Endo to bacteria have been well documented (Bissonnette et al., 1975; Cook et al., 1974; Dufour and Cabell, 1975; Klein and Wu, 1974; Knittel, 1975; Scheusner et al., 1971; Warseck et al., 1973). VRB on the other hand has demonstrated a capability of supporting the growth of much greater numbers of enteric bacteria than m-Endo. A comparison of counts of enterics obtained from both m-Endo and VRB is clearly indicative of the inhibitory effect of m-Endo. The inhibitory aspect of the media appears to be associated with dye concentrations in the formulation.

Dye concentrations of 1:100,000 will inhibit gram-positive bacteria while allowing gram-negative organisms to grow^{SP}. Obviously the 1.05 g/l (105 parts per 100,000) of basic fuchsin would be highly selective and severely limit the survival of enteric bacteria that do not readily ferment lactose. VRB on the other hand contains only 0.2 parts per 100,000 of crystal violet, a dye concentration that would be much less inhibitory to slow and non-lactose fermenting enteric genera.

When the dilution volumes of VRB and m-Endo plates selected for enteric enumeration are examined, it is apparent that dilution volumes were much smaller on VRB, yet this medium resulted in a significantly much higher count of enteric organisms. This also indicates that there may be far greater numbers of slow or non-lactose fermenting enteric tracers than there are lactose fermenting tracers. These results may also indicate that there may be more stressed or damaged enterics in the environment than are detectable with methods and media generally accepted as standard.

The results show that significantly fewer transfers of isolates from VRB grew on EMB than those transferred from m-Endo. In the case of EMB the high concentration (65 parts per 100,000) methylene blue dye is implicated. The few organisms already growing in the poisonous environment of Endo could withstand the similar concentration of dye in EMB.

The inhibitory effects of EMB on isolates taken from VRB may have been amplified during incubation. The environment of the incubator may have had a tendency to dehydrate the media. Dehydration would increase the dye concentration and thus increase its inhibitory action toward slow or non-lactose fermenting enterics.

Public health implications. Public health aspects relative to these results are clear. The large number of undetected enteric bacteria (with standard procedures) indicate there is the possibility of a potential public health hazard continuing undetected until it reaches infectious concentrations. This indicates that at the very least a critical review of currently accepted standards and procedures should be initiated.

Dilution effects. Examination of the dye envelope (Figure 15) indicates an overall broadening and amplitude attenuation as it proceeds from Sampling Point G to Sampling Point 2. It seemed that bacterial numbers and component distributions should follow the same trend. The lengthening and attenuation of bacterial numbers would have the same effect as dilution in decreasing the numbers of an incident population. Therefore decreases in enteric tracers may not be caused by broadening.

Component ratio concept. The graphic display (Figure 17) of the component genera of the lactose fermenting enteric tracers suggests that there could be no tracing of these bacteria much beyond Sampling Point G where tracer concentrations are a factor of ten times greater than background. Sampling Point 2 appears to be approaching the

limit of detectability for the tracers utilized. At this point, while still a detectable entity, they are of the same order of magnitude as background concentrations.

In applying the component ratio concept, it was found that it held to an amazing degree to G. This is especially significant in view of the enormous energy change, decreases in velocity, and dilution effects found in this region. This new concept may have great potential for future application with more sensitive methods.

Limits of the component ratio concept. Enteric tracer bacteria entering the lake via LVW are also indigenous to the lake and a residual population may be present at all times with some variation in the concentrations of the component genera. Therefore, to utilize enteric bacteria as tracers, lower numerical limits must be preestablished for detection and also for the entering concentration of tracer. The precept that individual genera or ratios must be intermittent should be applied. For example the data collected during this investigation indicate in one instance an entering tracer population whose component genera are in concentrations of 1 and $2 \times 10^2/100$ ml, and a background population of 3 and 7/100 ml in lake water. Thus, tracing beyond Sampling Point G could not be accomplished with any confidence

because tracer concentrations were diluted or broadened to the same order of magnitude as the residual population of the lake.

Although there is no control of the entering bacterial population limit, lower detection limits can be based on the residual population component concentrations. Figure 15 demonstrates that the detection cutoff for enteric tracers is reached when the individual components of the tracer fall below $3 \times 10^1/100$ ml (for an acceptable degree of confidence). Figure 17 also illustrates that extensive bacterial tracing in Las Vegas Bay would require a tremendous concentration of enteric organisms for them to be detectable above the residual population of the lake. If, however the enteric tracer is not indigenous to the lake and present intermittently in LVW, tracing could be accomplished with significantly lower enteric tracer concentrations.

During a period when very high concentrations of enteric bacteria were present in LVW (Table 26) the background (lake) concentrations of enteric tracer (Enterobacter colacae and Erwinea herbicola) were of the same order of magnitude as they were at the time of the dye study. Comparison of the component genera in the background population (Figures 16 and 17) demonstrates the

variability of the concentrations of the component genera within this population. This supports the hypothesis that total numbers of residual enteric bacteria remain relatively constant in the lake while numbers of component genera of the population show significant variability, an observation wighing against the "Component Ratio" tracing procedure. Note that in both cases the data was based on isolates taken from m-Endo media and the number of isolates per sample were not as numerous as was desired. However they did indicate the feasibility of utilizing component enteric genera to determine water distribution patterns. Obviously, isolation and identification of much greater numbers of enteric bacteria would bestow a greater degree of confidence on this kind of tracing, especially with the new technical concepts presented in this study.

The problems inherent in using enteric bacteria tracers are basically isolation of large numbers of enteric bacteria and rapid identification of those isolates. Both problems may be resolved by careful selection of media and refining present techniques.

Initial isolation and culture purification. The feasibility of initially isolating enteric bacteria on NB and replica plating to VRB has been demonstrated.

Examination of Table 25 indicates that the enteric population represent approximately three percent of the total bacterial population, however the actual mean number of enterics in the population may be significantly greater. Note that the mean of the enteric population is significantly lower than the standard deviation of the total bacteria count. This indicates that a fortified NB or tryptone glucose yeast extract (TGY) medium may result in higher counts of oxidase negative organisms. The results also indicate that VRB is capable of supporting the growth of a much greater number of enteric bacteria than m-Endo. This suggests that VRB may be a very good medium for both initial isolation and purification of enteric bacteria collected from natural environments.

Initial isolation of enteric organisms on non-selective media and incubating at temperatures below 30°C would have the advantage of allowing stressed and damaged bacteria time to repair themselves before being replica plated to differential media. The disadvantage of this procedure lies in the dilution volumes required for competent colony counting. For example, if the total bacterial concentration is 10^5 /ml, a dilution volume of sample of approximately 0.001 ml would be required to achieve a countable number of colonies per plate. Dilutions to this extent may eliminate

all the enterics associated with fecal pollution and significant number of the total enteric population. The results however indicate that the total enteric population would remain in traceable concentrations.

Identification of enteric bacteria. The Enterobacteriaceae are a family with closely related genera. Only few colony descriptors are available, with the exception of certain strains of Serratia marcescens there is a complete lack of chromogenesis within the family. Therefore descriptors for the component genera of the population must be based on the results of several biochemical tests.

Differentiation between Enterobacter aerogenes and Klebsiella pneumoniae is based on the lack of motility of K. pneumoniae and decarboxylation of ornithine by E. aerogenes. The reliability of these two characteristics is suspect because it has been demonstrated that some strains of E. aerogenes are non-motile and some strains are ornithine decarboxylase negative (American Public Health Association et al., 1971; Grimes, 1975).

In the majority of instances species identification of any member of the enteric group requires eleven or more biochemical descriptors (Edwards and Ewing, 1972; Johnson et al., 1975; McCoy and Seidler, 1973; Painter and Isenberg, 1973; Tomfohrde et. al., 1973), and in the case of Salmonella,

speciation can be achieved by specific antibody reactions only.

Identification of large numbers of enteric isolates would be very time consuming and costly if each individual isolate is treated separately. A possible alternative to this procedure would be replica plating a multidescrptor array from initial isolation plates. A method such as this would facilitate the simultaneous identification of a large number of enteric bacteria.

Replica plating a multidescrptor array poses certain technical problems that must first be resolved if it is to become a standard procedure. The primary problem is concerned with the transfer of discrete colonies to a series of plates containing the desceiptive media. The method generally used incorporates short nap velvet disks cut to the same dimensions as the isolation plates used (Lederberg and Lederberg, 1952). This method although satisfactory for two to three transfers has a tendency to distort the individual colonies and in some instances is responsible for cross contamination with adjacent colonies. These effects appear to be caused by the bending of the velvet nap. It appears that to continuously achieve discrete colonies on a series of plates a more rigid transfer applicator such as a densely bristled metal or nylon brush would be desirable.

Another problem involved in the transfer and maintenance of discrete colonies in a series of replica plates is the dryness of medium surfaces. A medium surface must be dry enough to inhibit the motility of the isolates, yet not so dry as to increase the concentration of medium constituents to the point where they become inhibitory or toxic to individual isolates.

Baseline grouping and a few confirmed identifications can be achieved with eight media (Bascomb et al., 1973; Cook et al., Edwards and Ewing, 1972; McCoy and Seidler, 1973; Painter and Isenberg 1973; Storey et al., 1974). Further description beyond baseline grouping would be dictated by individual groups and media would be made as needed.

Identification of Enteric Bacteria in Water Supplies

by

Sam Egdorf

INTRODUCTION

Standard Methods for the Examination of Water and Wastewater, 13th ed. (American Public Health Association et al., 1971) lists only two methods for presumptive and one method for confirmed identification of coliform bacteria. Both presumptive methods, membrane filter and most probable numbers, rely on lactose as the differential sugar. According to Cowan (1974), lactose fermentation is quite variable among the Enterobacteriaceae. Thus colonies counted and isolated may not be coliforms, but other enteric bacteria. For confirmation Standard Methods utilizes the IMViC tests, which are actually directed toward the characterization only of Escherichia coli, Enterobacter aerogenes, and Citrobacter freundii, or, 3 of the 11 enteric genera.

To test the extent and significance of the problem with presumptive procedures, a program of isolation and identification was initiated with environmental samples (Lake Mead and Las Vegas Wash) known to contain large numbers of enteric bacteria. To underscore the limitations of Standard Methods for confirmation, a slightly modified version of the multitest procedure advocated by Martin (1970) was applied to the same isolates subjected to IMViC testing.

MATERIALS AND METHODS

Water samples were collected in a three liter Van Dorn bottle sanitized with acetone-alcohol. The water column was sampled in 10 meter increments from surface to bottom at five sampling locations (Stations 1-5). The samples were placed on ice and transported to the laboratory for analysis. Duplicate dilutions of 100, 10, and 1 ml were filtered on membrane filters, treated with media, incubated, and counted according to Standard Methods procedures for fecal and total coliforms (American Public Health Association, et al., 1971).

Typical colonies were randomly selected from the filters, streaked on EMB agar, and incubated at 35°C until well-defined colonies appeared. Both typical and atypical coliform colonies on EMB were transferred to maintenance slants. When substantial growth was obtained on each of the slants, the individual isolates were gram-stained, inoculated into dextrose broth, and tested for oxidase and catalase capability. The organisms demonstrating characteristics of the Enterobacteriaceae as defined by Cowan (1974) were introduced to the IMViC and multitest procedures. ROCHE Improved Enterotubes were used for the latter.

After all biochemical tests were complete, the organisms were identified according to schemes for both procedures. No additional tests were performed on organisms not identified by the IMViC method. Rhamnose and raffinose fermentations were required in some instances to supplement the 11 biochemical tests of the multitest procedure.

RESULTS

The results of the comparison demonstrates the inadequacy of the IMViC method of identification, which resulted in relatively low numbers of identified organisms (Table 27). On the other hand the multitest procedure resulted in 100 percent identification of all organisms introduced into the system.

For example isolate 1, identified as Escherichia coli Variety I by the IMViC procedure, demonstrated characteristics that were actually those of Klebsiella spp. as indicated by biochemical reactions, lack of motility and the presence of a capsule. The results of the reactions of the IMViC tests together with the multitest procedure, indicated that the organism was either Klebsiella pneumoniae, type 3, or Klebsiella ozaenae.

Table 27. Identity of isolates by IMViC and Multitest procedures.

Isolate #	Identity By IMViC	Identity By Multitest
1	Escherichia coli Var. I	Klebsiella spp.
2	Enterobacter aerogenes Var. I	Klebsiella spp.
3	Escherichia coli Var. I	Escherichia coli
4	Enterobacter aerogenes Var. I	Klebsiella spp.
5	NOT IDENTIFIED	Escherichia coli
6	NOT IDENTIFIED	Shigella spp.
7	Enterobacter aerogenes Var. I	Enterobacter cloacae
8	NOT IDENTIFIED	Escherichia coli
9	NOT IDENTIFIED	Escherichia coli
10	Enterobacter aerogenes Var. I	Klebsiella spp.
11	NOT IDENTIFIED	Serratia marcescens
12	Enterobacter aerogenes Var. I	Enterobacter cloacae
13	NOT IDENTIFIED	Escherichia coli
14	NOT IDENTIFIED	Escherichia coli
15	NOT IDENTIFIED	Klebsiella spp.
16	NOT IDENTIFIED	Klebsiella spp.
17	Escherichia coli Var. II	Escherichia coli
18	Enterobacter aerogenes Var. II	Enterobacter cloacae

Isolates 2, 4 and 10 were identified as Enterobacter aerogenes Variety I by IMViC but the multitest procedure confirmed their identity as Klebsiella spp. The presence of ornithine decarboxylase was the only biochemical test differentiating E. aerogenes and K. pneumoniae; the lack of motility may be another identifying characteristic, but, according to Johnson (1975), these characteristics are not sufficient to differentiate genera, and, because they are so closely related, they should be combined into a single genus that would accommodate both motile and nonmotile forms.

Isolates 7, 12 and 18 were identified as E. aerogenes Varieties I, I and II, respectively, by IMViC. On the basis of rhamnose fermentation and lack of raffinose fermentation the multitest indicated that E. aerogenes Varieties I and II were actually E. cloacae, a different species.

Of the 18 isolates utilized in this comparison, only 50 percent were identified by the Standard Method procedure (IMViC), and of those identified by this method, only two were correct by current standards for nomenclature. The multitest procedure identified all of the isolates to a great degree of confidence with the exception of isolate 1. This particular organism would have required a much more

detailed biochemical series of tests to establish whether it was K. pneumoniae or K. ozaenae. Table 1 contains organism identification for both the IMViC, and multimedia systems.

DISCUSSION

The multitest procedure employed in this study greatly facilitated the identification of the isolated enterics. This was pointed out by Leers (1973) who stated that more than 92 percent of the enteric bacteria could be identified within 24 hours without additional tests. Actually, we found that all the isolates could be identified to genus within 24 hours, but further tests were required to apply epithets to species of Klebsiella, Enterobacter, Citrobacter, Shigella, and Salmonella. Although not shown in the rapid screening chart, Yersinia spp. can also be identified by the use of this multitest method of identification.

In all instances Cowan's (1974) methods should be utilized in conjunction with the rapid screening chart because isolates do not always demonstrate a classic identification profile, and, unless the technician examines all possibilities, the probability of misidentification is always present.

The multitest system, which perhaps not the ultimate answer for enteric identification, appears to be the

best interim procedure we have at this time. Similar methods have been used in clinical bacteriology, computer assisted taxonomy, and numerical taxonomy (Finlayson and Gibbs, 1974; Friedman et al., 1973; Johnson et al., 1975; Martin, 1970; Oliver and Cowell, 1974; Shewane et al., 1960).

Standard Methods procedures, on the other hand, seem burdened with inherent possibilities for error, with lactose fermentation with gas formation within 24⁺ 2 hours at 44.5°C the principle basic problem. Only certain biotypes of E. coli and C. freundii exhibit the capability. Also there is some disagreement concerning E. aerogenes. It has been shown that E. aerogenes capable of fermenting lactose with gas production at 44.5°C appears to be from soil or vegetation free from fecal contamination, while the other biotype, negative for this reaction, appears to be of fecal origin (Cowan, 1974). This is in direct contradiction with Standard Methods.

For the above reason we feel that the multitest method of identification should be considered for the next revision of Standard Methods. This would allow substitution of identification of the enteric bacteria actually present for the rather suspect current "coliform" procedures. Obviously, lactose in present presumptive media would have to be replaced by dextrose, the sugar common to all Enterobacteriaceae.

Water Quality Standards
North Shore Road

Environmental Protection Agency standards for Las Vegas Wash at North Shore Road and parameter determined in 1975 are summarized in Table 28. Temperature, pH, dissolved oxygen, methylene blue-active substances, and total free chlorine are within the standards.

MBAS, which is a measurement of the surface-active agents or surfactants in synthetic detergents, was below 1 mg/l, the level at which frothing occurs. Although levels were below 1 mg/l, frothing did occasionally occur at North Shore Road, therefore, levels may have been higher than test results indicated. Both organic and inorganic compounds interfere with the determination of surfactants when using the methylene blue technique although positive errors are more common than negative errors. Amines can cause a negative error and may have resulted in lower values of surfactants than were actually present.

Biological oxygen demand (BOD₅), chemical oxygen demand (COD), phosphorous, suspended solids, turbidity, and coliforms were higher than E.P.A. standards.

Table 28. E.P.A. water quality standards for Las Vegas Wash at North Shore Road and values found in 1975.

Parameter	Units	EPA Standards	1975	
			Mean	Range
Temperature	oc	<32	18.2	10.0 - 23.5
pH	units	6.5 - 8.5	8.1	7.0 - 8.4
Dissolved oxygen	mg/l	≥5.0	8.7	6.5 - 11.8
BOD ₅	mg/l	10.0	9.9	0 - 40.8
COD	mg/l	40.0	85	33.3 - 148.0
Suspended solids	mg/l	2.0	470	113 - 1770
MBAS	mg/l	0.8	0.1	0.02 - 0.24
Phosphorous as P	mg/l	0.5	4.53	3.88 - 5.08
Turbidity	JTU	1.0	99	28 - 325
Fecal coliforms	-	<2/100 ml.	344	0 - 1700

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Appendixes

Appendix I: Nutrient concentrations in mg/l at stations 1-6, 1975 and 1976.
 Total-P = total phosphorus; Diss-P = dissolved phosphorus; NH₃-N = ammonia nitrogen; NO₂ + NO₃ = nitrite plus nitrate nitrogen; TKN = total kjeldahl nitrogen; Total-ALK = total alkalinity.

STATION 1							
Date	Depth	Total-P	Diss-P	NH ₃ -N	NO ₂ +NO ₃	TKN	Total-ALK
28 Apr	0	4.63	4.33	0.03	7.28	0.8	258
16 May	0		5.07	0.24	8.40	1.1	-
28 May	0	4.74	4.69	0.11	12.10	1.0	252
17 Jun	0	4.68	4.32	0.09	10.20	1.2	204
27 Jun	0	4.84	3.88	0.08	10.30	1.0	272
21 Jul	0	4.12	3.12	1.68	13.30	2.9	340
31 Jul	0	4.56	4.73	0.23	8.80	0.8	316
28 Aug	0	3.99	3.63	0.14	1.04	1.1	119
17 Sept	0	5.08	0.41	0.63	6.78	3.0	280
2 Oct	0	3.88	3.87	0.04	1.00	1.2	290
16 Oct	0	4.83	4.55	0.02	14.30	1.2	290
30 Oct	0	4.64	4.60	0.64	10.16	0.2	248
20 Nov	0	4.42	4.16	0.44	11.10	0.8	278
22 Dec	0	4.46	4.51	1.60	13.80	2.0	194
29 Jan	0	4.70	4.50	3.40	10.40	2.9	300
19 Feb	0	4.40	4.40			2.5	

Appendix I (continued)

STATION 2

Date	Depth	Total-P	Diss-P	NH ₃ -N	NO ₂ +NO ₃	TKN	Total ALK
28 Apr	0	0.087	0.017	0.06		0.5	130
	10	1.37	1.21	0.03	0.14	0.5	154
16 May	0	0.14	0.11	0.07	0.16	0.7	126
	10	2.50	2.17	0.15	4.34	0.9	170
28 May	0	0.05	0.01	0.04	0.03	0.5	127
	10	1.24	0.01	0.09	2.14	0.8	147
17 Jun	0	0.07	0.07	0.04	0.09	0.6	114
	10	3.00	1.66	0.12	2.93	1.1	160
27 Jun	0	0.14	0.09	0.02	0.07	0.8	122
	10	2.10	1.22	0.06	3.43	1.1	156
21 Jul	0	0.05	0.04	0.04	0.18	0.9	94
	10	0.30	0.27	0.02	0.66	0.6	120
31 Jul	0	0.11	0.06	0.02	0.06	0.6	103
	10	1.08	1.29	0.04	2.40	0.6	137
19 Aug	0	0.17	0.05	0.04	0.04	0.7	91
	10	0.19	0.06	0.04	0.29	0.5	97
28 Aug	0	0.06	0.05	0.02	0.16	0.6	136
	10	0.09	0.06	0.02	0.08	0.4	115
17 Sept	0	0.061	0.037	0.03	0.07	0.5	97
	10	0.068	0.049	0.03	0.08	0.4	108

Appendix

STATION

<u>Date</u>	<u>Depth</u>	<u>Total-P</u>	<u>Diss-P</u>
2 Oct	0	0.061	0.034
	10	0.050	0.021
16 Oct	0	0.096	0.048
	10	0.084	0.043
30 Oct	0	0.100	0.069
	10	0.079	0.029
20 Nov	0	0.054	0.046
	10	0.124	0.076
22 Dec	0	0.045	
	10	0.033	
29 Jan	0	0.048	0.048
	10	0.049	0.021
19 Feb	0	0.055	0.028
	10	0.052	0.023

I (continued)

2 (continued)

<u>NH₃-N</u>	<u>NO₂+NO₃</u>	<u>TKN</u>	<u>Total ALK</u>
0.02	0.17	0.8	105
0.02	0.05	0.4	109
0.02	0.07	0.6	111
0.02	0.08	0.4	110
0.02	0.32	0.4	236
0.02	0.10	0.2	114
0.04	0.45	0.4	133
0.04	0.45	0.3	131
0.06	0.39	0.5	137
0.03	0.30	0.4	136
0.05	0.34	0.9	133
0.04	0.35	0.4	136
		0.5	
		0.4	

Appendix I (continued)

STATION 3

Date	Depth	Total-P	Diss-P	NH ₃ -N	NO ₂ +NO ₃	TKN	Total ALK
28 Apr	0	0.038	0.014	0.03	0.14	0.5	132
	10	0.031	0.007	0.03	0.20	0.3	133
	20	0.041	0.031	0.04	0.33	0.2	133
	30	0.068	0.059	0.04	0.42	0.2	134
	40	0.156	0.139	0.04	0.54	0.2	136
16 May	0	0.026	0.021	0.09	0.13	0.4	130
	10	0.100	0.010	0.11	0.12	0.4	138
	20	0.028	0.015	0.08	0.24	0.3	139
	30	0.211	0.192	0.08	0.52	0.3	142
	40	0.143	0.132	0.06	0.52	0.3	142
28 May	0	0.019	0.007	0.04	0.14	0.2	132
	10	0.039	0.005	0.04	0.11	0.2	132
	20	0.130	0.118	0.12	0.30	0.2	137
	30	0.107	0.095	0.10	0.33	0.2	136
	40	0.179	0.165	0.09	0.51	0.2	140
17 Jun	0	0.024	0.023	0.038	0.054	0.42	117
	10	0.024	0.011	0.05	0.14	0.5	131
	20	0.039	0.007	0.04	0.10	0.4	127
	30	0.175	0.160	0.06	0.51	0.3	139
	40	0.107	0.099	0.04	0.46	0.3	139
27 Jun	0	0.061	0.031	<.02	0.03	0.6	120
	10	0.032	0.013	<.02	0.07	0.4	128
	20	0.091	0.076	0.04	0.38	0.4	137
	30	0.080	0.068	<.02	0.43	0.3	146
	40	0.078	0.068	<.02	0.46	0.3	143
31 Jul	0	0.046	0.019	<.02		0.4	106
	10	0.078	0.053	<.02		0.4	123
	20	0.077	0.055	<.02		0.2	136
	30	0.148	0.116	<.02		0.3	137
	40	0.114	0.093	<.02		0.2	138

Appendix

STATION

Date	Depth	Total-P	Diss-P
19 Aug	0	0.023	0.016
	10	0.036	0.011
	20	0.092	0.087
	30	0.110	0.105
	40	0.091	0.084
28 Aug	0	0.029	0.011
	10	0.021	0.006
	20	0.227	0.219
17 Sept	0	0.018	0.009
	10	0.024	0.006
	20	0.132	0.121
	30	0.090	0.087
	40	0.86	0.079
2 Oct	0	0.034	0.009
	10	0.028	0.007
	20	0.409	0.395
	30	0.109	0.101
	40	0.145	0.130
16 Oct	0	0.047	0.019
	10	0.050	0.020
	20	0.156	0.129
	30	0.070	0.056
	40	0.075	0.063
30 Oct	0	0.044	0.019
	10	0.048	0.018
	20	0.052	0.020
	30	0.124	0.085
	40	0.059	0.018

I (continued)

3 (continued)

<u>NH₃-N</u>	<u>NO₂+NO₃</u>	<u>TKN</u>	<u>Total ALK</u>
<.02	0.33	0.4	105
<.02	0.06	0.3	104
<.02	0.41	0.4	132
<.02	0.56	0.4	136
<.02	0.54	0.4	133
<.02	<.02	0.3	116
<.02	<.02	0.3	128
0.05	0.52	0.3	123
<.02	0.02	0.2	104
<.02	<.02	0.2	106
<.02	0.46	<.2	143
<.02	0.48	<.2	147
<.02	0.48	0.2	146
<.02	0.03	0.4	112
<.02	0.02	0.4	112
<.02	0.96	0.6	141
<.02	0.45	0.4	146
<.02	0.46	0.4	149
0.03	0.05	0.3	110
<.02	0.05	0.3	111
0.09	0.44	0.4	127
0.06	0.44	0.2	136
0.05	0.44	<.2	135
0.03	0.10	0.2	116
0.03	0.10	0.2	115
0.03	0.10	0.2	116
0.06	0.25	0.4	115
0.03	0.10	0.2	112

Appendix

STATION

Date	Depth	Total-P	Diss-P
20 Nov	0	0.033	0.014
	10	0.034	0.011
	20	0.034	0.010
	30	0.031	0.010
	40	0.064	0.035
22 Dec	0	0.025	0.010
	10	0.023	0.009
	20	0.021	0.009
	30	0.019	0.008
	40	0.031	0.002
29 Jan	0	0.031	0.013
	10	0.035	0.013
	20	0.028	0.013
	30	0.029	0.013
	40	0.033	0.016
19 Feb	0	0.046	0.009
	10	0.040	0.009
	20	0.041	0.011
	30	0.041	0.013
	40	0.038	0.014

I (continued)

3 (continued)

<u>NH₃-N</u>	<u>NO₂+NO₃</u>	<u>TKN</u>	<u>Total ALK</u>
0.03	0.19	0.2	135
0.03	0.18	<.2	132
0.04	0.18	<.2	132
0.03	0.18	<.2	131
0.04	0.23	<.2	131
0.03	0.27	0.2	136
0.03	0.26	0.2	136
0.03	0.25	0.2	134
0.03	0.25	0.2	134
0.04	0.28	0.2	134
0.03	0.32	0.2	137
0.03	0.31	0.2	135
0.03	0.31	0.2	134
0.03	0.31	0.2	138
0.03	0.31	0.2	136
		0.4	
		0.3	
		0.3	
		0.2	
		0.2	

Appendix I (continued)

STATION 4

Date	Depth	Total-P	Diss-P	NH ₃ -N	NO ₂ +NO ₃	TKN	Total ALK
28 Apr	0	0.017	0.009	0.03	0.24	0.3	133
	10	0.035	0.025	0.03	0.26	0.3	134
	20	0.017	0.008	0.03	0.33	<.2	131
	30	0.048	0.041	0.03	0.42	<.2	132
	90	0.021	0.022	0.02	0.44	<.2	134
16 May	0	0.017	0.010	0.06	0.17	0.4	138
	10	0.023	0.005	0.06	0.22	0.3	140
	20	0.015	0.005	0.06	0.30	0.2	138
	30	0.016	0.008	0.04	0.36	0.2	140
	90	0.030	0.250	0.04	0.44	<.2	140
28 May	0	0.016	0.008	0.05	0.16	<.2	135
	10	0.018	0.004	0.04	0.18	<.2	136
	20	0.019	0.012	0.08	0.23	<.2	137
	30	0.046	0.039	0.066	0.30	<.2	137
	40	0.046	0.040	0.04	0.38	<.2	139
	90	0.030	0.026	0.03	0.42	<.2	138
17 Jun	0	0.017	0.010	0.04	0.10	0.4	120
	10	0.018	0.004	0.04	0.20	0.3	131
	20	0.012	0.003	0.05	0.25	0.3	135
	30	0.022	0.007	<.02	0.33	0.3	136
	40	0.028	0.017	<.02	0.38	0.3	137
	90	0.014	0.003	<.02	0.08	0.4	120
27 Jun	0	0.015	0.009	<.02	0.04	0.4	125
	10	0.020	0.007	<.02	0.04	0.5	117
	20	0.029	0.015	0.05	0.29	0.3	135
	30	0.023	0.016	<.02	0.35	0.3	134
	40	0.027	0.019	<.02	0.38	0.3	135
	90	0.039	0.023	<.02	0.42	0.3	137

Appendix I

STATION 4

Date	Depth	Total-P	Diss-P
31 Jul	0	0.017	0.009
	10	0.017	0.006
	20	0.014	0.004
	30	0.027	0.013
	40	0.037	0.021
	90	0.041	0.017
19 Aug	0	0.015	0.004
	10	0.014	0.002
	20	0.011	0.002
	30	0.030	0.024
	40	0.018	0.012
	90	0.056	0.027
28 Aug	0	0.016	0.011
	10	0.008	0.004
	20	0.008	0.003
17 Sept	0	0.016	0.015
	10	0.013	0.007
	20	0.007	0.005
	30	0.014	0.010
	40	0.015	0.011
	90	0.028	0.023
2 Oct	0	0.015	0.009
	10	0.019	0.006
	20	0.019	0.010
	30	0.017	0.010
	40	0.024	0.014
	90	0.036	0.028

(continued)

(continued)

<u>NH₃-N</u>	<u>NO₂+NO₃</u>	<u>TKN</u>	<u>Total ALK</u>
<.02	0.04	0.3	112
<.02	0.03	0.3	116
<.02	0.34	0.3	138
<.02	0.41	0.2	137
<.02	0.43	0.2	138
<.02	0.48	0.3	140
<.02	0.04	0.4	109
<.02	0.03	0.4	111
<.02	0.31	0.3	134
<.02	0.46	0.3	136
<.02	0.43	0.3	134
<.02	0.46	0.4	136
<.02	0.02	0.3	143
<.02	0.02	0.3	116
<.02	0.26	<.2	124
0.02	0.02	<.2	101
0.02	<.02	0.3	105
0.02	0.32	0.2	137
0.02	0.39	<.2	134
0.02	0.40	<.2	134
0.02	0.42	<.2	136
<.02	0.04	0.4	117
<.02	0.02	0.4	116
<.02	0.29	0.4	145
<.02	0.40	0.4	148
<.02	0.43	0.6	147
<.02	0.45	0.6	150

Appendix I (continued)

STATION 4 (continued)

Date	Depth	Total-P	Diss-P	NH ₃ -N	NO ₂ +NO ₃	TKN	Total ALK
16 Oct	0	0.022	0.009	<.02	0.04	0.2	115
	10	0.018	0.006	<.02	0.03	<.2	115
	20	0.014	0.006	<.02	0.36	<.2	138
	30	0.008	0.004	<.02	0.39	<.2	137
	40	0.017	0.011	<.02	0.42	<.2	136
	90	0.037	0.23	<.02	0.43	<.2	139
30 Oct	0	0.024	0.005	0.02	0.13	<.2	118
	10	0.021	0.004	<.02	0.13	<.2	117
	20	0.022	0.003	<.02	0.13	<.2	117
	30	0.146	0.126	<.02	0.61	<.2	130
	40	0.014	0.013	<.02	0.44	<.2	129
	90	0.031	0.021	<.02	0.44	<.2	132
20 Nov	0	0.020	0.006	0.02	0.20	<.2	129
	10	0.019	0.004	0.02	0.20	<.2	131
	20	0.021	0.005	0.02	0.20	<.2	127
	30	0.032	0.011	0.03	0.20	<.2	127
	40	0.077	0.061	0.04	0.45	<.2	135
	90	0.032	0.023	0.02	0.45	<.2	142
22 Dec	0	0.019	0.009	0.03	0.27	0.2	137
	10	0.017	0.008	0.03	0.27	0.2	135
	20	0.025	0.010	0.04	0.26	0.3	136
	30	0.019	0.010	0.05	0.26	0.2	133
	40	0.021	0.009	0.04	0.26	0.2	135
	90	0.025	0.026	0.02	0.44	0.3	143
29 Jan	0	0.025	0.014	0.03	0.31	0.2	136
	10	0.023	0.014	0.03	0.32	0.2	136
	20	0.020	0.014	0.02	0.32	0.2	134
	30	0.020	0.014	0.02	0.31	0.2	134
	40	0.020	0.012	0.02	0.31	0.2	137
	90	0.045	0.034	0.04	0.36	0.3	140

Appendix I

Station 4

Date	Depth	Total-P	Diss-P
19 Feb	0	0.028	0.007
	10	0.030	0.006
	20	0.019	0.015
	30	0.022	0.012
	40	0.021	0.008
	90	0.076	0.019

(continued)

(continued)

<u>NH₃-N</u>	<u>NO₂+NO₃</u>	<u>TKN</u>	<u>Total ALK</u>
		0.2	
		0.3	
		0.2	
		0.2	
		0.2	
		0.3	

Appendix I (continued)

STATION 5

Date	Depth	Total-P	Diss-P	NH ₃ -N	NO ₂ +NO ₃	TKN	Total ALK
May 9	0	0.027	0.025	0.04	0.24	0.4	142
	5	0.036	0.009	0.03	0.199	0.4	140
	10	0.023	0.009	0.04	0.22	0.5	139
	15	0.018	0.004	0.03	0.26	0.4	138
	20	0.021	0.008	0.07	0.31	0.9	137
	30	0.013	0.005	0.03	0.34	0.4	136
	40	0.024	0.016	0.02	0.39	0.4	137
	45	0.026	0.019	0.02	0.40	0.3	139
17 Jun	0	0.013	<.002	<.02	0.08	0.5	123
	5	0.012	<.002	<.02	0.14	0.3	128
	10	0.014	<.002	<.02	0.16	0.3	133
	15	0.011	<.002	0.04	0.24	0.3	138
	20	0.010	<.002	0.06	0.24	0.3	136
	30	0.012	<.002	<.02	0.34	0.2	137
	40	0.026	0.015	<.02	0.38	<.2	135
2 Jul	0	0.018	0.004	0.03	<.02	0.4	125
	5	0.020	0.003	0.02	0.037	0.45	124
	10	0.023	0.003	<.02	0.023	0.47	121
	15	0.024	0.007	<.02	0.13	0.5	132
	20	0.017	0.003	0.04	0.28	0.4	141
	30	0.017	0.005	<.02	0.35	0.4	139
	40	0.018	0.009	<.02	0.37	0.3	140
18 Jul	0	0.007	0.004	<.02	0.04	0.2	107
	5	0.006	0.003	<.02	<.02	0.2	107
	10	0.006	0.002	<.02	0.05	0.2	114
	15	0.007	<.002	<.02	0.17	0.2	123
	20	0.006	0.003	<.02	0.23	0.2	129
	30	0.004	0.004	<.02	0.37	<.2	131
	40	0.009	0.008	<.02	0.39	<.2	132
	45	0.014	0.014	<.02	0.41	<.2	133

Appendix I

STATION 5

Date	Depth	Total-P	Diss-P
28 Aug	0	0.011	0.004
	5	0.010	0.003
	10	0.009	0.003
	15	0.003	.002
	20	0.007	0.004
	30	0.006	0.006
	40	0.010	0.010
2 Oct	0	0.014	0.005
	5	0.012	0.004
	10	0.017	0.009
	15	0.016	0.005
	20	0.012	0.005
	30	0.010	0.004
	40	0.012	0.005
	45	0.013	0.007
16 Oct	0	0.015	0.005
	5	0.018	0.004
	10	0.016	0.003
	15	0.015	0.003
	20	0.012	0.002
	30	0.012	0.004
	40	0.010	0.004
	45	0.013	0.007
30 Oct	0	0.017	0.006
	10	0.017	0.005
	20	0.017	0.003
	30	0.009	0.003
	40	0.010	0.004

(continued)

(continued)

<u>NH₃-N</u>	<u>NO₂+NO₃</u>	<u>TKN</u>	<u>Total ALK</u>
<.02	0.02	0.2	146
<.02	0.02	0.3	120
<.02	0.02	0.2	118
<.02	0.24	0.2	125
<.02	0.39	0.2	148
<.02	0.40	0.2	148
<.02	0.40	0.2	148
<.02	0.04	0.8	117
<.02	0.03	0.8	117
<.02	0.02	0.2	102
0.02	<.02	0.2	105
<.02	0.31	<.2	137
<.02	0.37	<.2	135
<.02	0.39	<.2	137
<.02	0.39	<.2	138
<.02	0.04	0.2	113
<.02	0.03	0.2	114
<.02	0.03	<.2	116
<.02	0.03	0.2	114
<.02	0.28	0.2	113
<.02	0.40	<.2	136
<.02	0.41	<.2	135
<.02	0.40	<.2	136
<.02	0.14	<.2	116
<.02	0.14	<.2	119
<.02	0.14	<.2	119
<.02	0.40	<.2	137
<.02	0.41	<.2	136

Appendix I (continued)

STATION 5 (continued)

Date	Depth	Total-P	Diss-P	NH ₃ -N	NO ₂ +NO ₃	TKN	Total Alk
20 Nov	0	0.022	0.009	0.03	0.22	<.2	132
	5	0.020	0.007	0.02	0.21	<.2	137
	10	0.020	0.006	0.02	0.20	<.2	133
	15	0.022	0.006	0.02	0.21	<.2	133
	20	0.021	0.006	0.04	0.21	<.2	131
	30	0.021	0.007	0.03	0.21	<.2	138
	40	0.035	0.021	0.04	0.39	<.2	135
	45	0.014	0.009	0.02	0.44	<.2	137
23 Dec	0	0.018	0.011	0.03	0.28	0.2	135
	10	0.019	0.009	0.03	0.27	0.2	134
	20	0.018	0.009	0.02	0.28	0.2	129
	30	0.018	0.008	0.02	0.28	0.2	132
	40	0.019	0.013	0.03	0.28	0.2	138
29 Jan	0	0.030	0.010	0.02	0.28	2	138
	5	0.029	0.009	0.02	0.28	2	140
	10	0.032	0.008	0.02	0.29	2	138
	15	0.023	0.009	0.02	0.29	2	137
	20	0.022	0.016	0.02	0.30	4	140
	30	0.032	0.011	0.02	0.31	2	141
	40	0.030	0.017	0.03	0.33	3	139
	45	0.031	0.023	0.04	0.34	2	139
19 Feb	0	0.028	0.012			2	
	5	0.026	0.008			2	
	10	0.025	0.005			3	
	15	0.028	0.005			2	
	20	0.020	0.005			3	
	30	0.018	0.008			2	
	40	0.017	0.009			2	
	45	0.018	0.010			2	

Appendix I (continued)

STATION 6

Date	Depth	Total-P	Diss-P	NH ₃ -N	NO ₂ +NO ₃	TKN	Total ALK
28 Apr	0	0.019	0.005	0.03	0.25	<.2	132
	10	0.022	0.005	0.04	0.28	<.2	132
	20	0.016	0.010	0.04	0.33	<.2	130
	30	0.019	0.014	0.03	0.38	<.2	140
	90	0.026	0.023	0.02	0.44	<.2	174
	130	0.027	0.021	<.02	0.44	<.2	143
16 May	0	0.016	0.006	0.05	0.17	0.4	137
	10	0.022	0.007	0.07	0.19	0.4	142
	20	0.022	0.007	0.07	0.19	0.3	142
	30	0.016	0.009	0.04	0.36	<.2	140
	90	0.026	0.022	0.03	0.44	<.2	143
	130	0.029	0.023	0.04	0.45	<.2	142
28 May	0	0.012		0.02	0.17	<.2	136
	10	0.014	0.006	0.06	0.17	<.2	135
	20	0.010	0.004	0.07	0.25	<.2	135
	30	0.010	0.006	0.06	0.30	<.2	137
	40	0.020	0.016	0.03	0.36	<.2	136
	90	0.025	0.021	0.03	0.41	<.2	137
	130	0.027	0.022	0.02	0.42	<.2	138
27 Jun	0	0.011	0.003	<.02	0.07	0.3	122
	10	0.012	0.002	<.02	0.061	0.37	122
	20	0.026	0.015	<.03	0.31	0.4	140
	30	0.011	0.004	<.02	0.32	0.3	138
	40	0.13	0.005	<.02	0.37	0.3	141
	90	0.021	0.015	0.03	0.41	0.3	144
	130	0.028	0.020	<.02	0.43	0.3	141
31 Jul	0	0.016	0.006	<.02	0.04	0.4	118
	10	0.016	0.005	<.02	0.03	0.4	119
	20	0.012	0.004	<.02	0.34	0.5	137
	30	0.011	0.004	<.02	0.38	0.3	139
	40	0.013	0.004	<.02	0.40	0.3	140
	90	0.024	0.009	<.02	0.43	0.3	135
	130	0.033	0.015	<.02	0.44	0.3	136

Appendix I (continued)

STATION 6 (continued)

Date	Depth	Total-P	Diss-P	NH ₃ -N	NO ₂ +NO ₃	TKN	Total ALK
19 Aug	0	0.018	0.005	<.02	0.04	0.4	113
	10	0.012	0.002	<.02	0.02	0.2	114
	20	0.009	0.002	<.02	0.30	0.2	135
	30	0.009	0.002	<.02	0.39	0.2	136
	40	0.008	0.002	<.02	0.40	0.2	136
	90	0.019	0.015	<.02	0.42	0.2	138
	130	0.025	0.020	<.02	0.43	0.3	138
28 Aug	0	0.006	0.003	<.02	0.02	<.2	148
	10	0.006	0.003	<.02	0.02	<.2	123
	20	0.003	<.002	<.02	0.32	<.2	124
17 Sept	0	0.010	0.006	<.02	0.02	0.2	105
	10	0.007	0.003	0.02	<.02	0.2	104
	20	0.003	<.002	0.02	0.36	<.2	140
	30	0.003	<.002	<.02	0.38	<.2	136
	40	0.006	<.002	<.02	0.38	<.2	134
	90	0.009	0.006	0.02	0.40	0.2	134
	130	0.022	0.019	<.02	0.41	<.2	133
2 Oct	0	0.012	0.003	<.02	<.02	0.3	111
	10	0.012	0.002	<.02	<.02	0.2	111
	20	0.010	0.010	<.02	0.18	<.2	135
	30	0.013	0.007	<.02	0.38	<.2	142
	40	0.009	0.004	<.02	0.38	<.2	139
	90	0.013	0.007	<.02	0.39	<.2	142
16 Oct	0	0.017	0.004	<.02	0.04	0.2	112
	10	0.016	0.005	<.02	0.04	0.2	115
	20	0.012	0.003	0.04	0.17	<.2	125
	30	0.012	0.007	0.04	0.39	0.2	136
	40	0.009	0.006	0.03	0.41	0.2	135
	90	0.011	0.013	0.04	0.42	0.2	136
	130	0.020	0.023	0.05	0.43	<.2	137

Appendix I (continued)

STATION 6 (continued)

Date	Depth	Total-P	Diss-P	NH ₃ -N	NO ₂ +NO ₃	TKN	Total ALK
30 Oct	0	0.019	0.004	<.02	0.12	<.2	120
	10	0.021	0.004	<.02	0.11	<.2	122
	20	0.019	0.003	<.02	0.11	<.2	120
	30	0.012	0.004	<.02	0.41	<.2	136
	40	0.011	0.005	<.02	0.38	<.2	133
	90	0.020	0.013	<.02	0.40	<.2	134
	130	0.015	0.005	<.02	0.35	<.2	131
20 Nov	0	0.021	0.007	0.03	0.21	<.2	124
	10	0.024	0.006	0.03	0.20	<.2	126
	20	0.023	0.006	0.03	0.20	<.2	125
	30	0.023	0.007	0.03	0.20	<.2	123
	40	0.029	0.017	0.03	0.33	<.2	129
	90	0.014	0.09	0.03	0.42	<.2	136
	130	0.037	0.026	0.02	0.44	<.2	141
22 Dec	0	0.022	0.008	0.02	0.28	0.2	132
	10	0.021	0.008	0.02	0.27	0.2	131
	20	0.018	0.008	0.02	0.27	0.4	133
	30	0.018	0.008	0.02	0.28	0.3	133
	40	0.020	0.009	0.03	0.27	0.3	133
	90	0.013	0.008	0.02	0.43	0.2	142
	130	0.031	0.020	0.02	0.46	0.2	141
29 Jan	0	0.024	0.014	0.02	0.32	0.3	137
	10	0.021	0.010	0.02	0.32	0.3	138
	20	0.021	0.010	0.02	0.33	0.2	135
	30	0.021	0.010	0.02	0.32	0.2	134
	40	0.022	0.010	0.02	0.32	0.2	137
	90	0.021	0.017	0.02	0.42	0.2	143
	130	0.034	0.021	0.02	0.44	0.2	146
19 Feb	0	0.026	0.008			0.2	
	10	0.018	0.006			0.2	
	20	0.018	0.006			0.2	
	30	0.018	0.005			0.4	

Appendix II. Dominant phytoplankton in Las Vegas

Date	Station 2	#/ml	Station 3	#/ml
28 Apr.	Chlamydomonas	149	Oocystis	38
	Carteria	107	Glenodinium	13
	Glenodinium	74	Carteria	12
	Oocystis	41	Fragilaria	7
	Total	418	Total	105
16 May	Cyclotella	228	Cyclotella	36
	Platymonas	69	Platymonas	30
	Chlamydomonas	41	Oocystis	21
	Sphaerocystis	23	Sphaerocystis	10
	Total	449	Total	139
28 May	Fragilaria	192	Chlorella	53
	Chlorella	135	Fragilaria	42
	Platymonas	84	Oocystis	27
	Chlamydomonas	77	Certium	12
	Total	600	Total	177
17 June	Fragilaria	112	Fragilaria	82
	Franceia	105	Oocystis	19
	Carteria	82	Carteria	16
	Glenodinium	63	Cyclotella	10
	Total	479	Total	166
27 June	Cyclotella	355	Cyclotella	74
	Glenodinium	229	Carteria	72
	Carteria	141	Platymonas	39
	Platymonas	75	Glenodinium	33
	Total	907	Total	317

Bay and Boulder Basin 1975-76.

Station 4	#/ml	Station 6	#/ml
Oocystis	31	Oocystis	28
Carteria	10	Carteria	13
Sphaerocystis	8	Shaerocystis	12
Fragilaria	7	Diatoma	5
Total	82	Total	87
Oocystis	31	Oocystis	29
Sphaerocystis	21	Sphaerocystis	21
Fragilaria	15	Fragilaria	12
Chlamydomonas	5	Ceratium	5
Total	85	Total	89
Oocystis	36	Oocystis	51
Fragilaria	27	Fragilaria	17
Ceratium	15	Sphaerocystis	12
Cynedra	5	Ceratium	5
Total	90	Total	97
Fragilaria	43		
Sphaerocystis	31		
Ceratium	8		
Oocystis	5		
Total	108		
Cyclotella	27	Fragilaria	28
Fragilaria	25	Sphaerocystis	25
Carteria	15	Cyclotella	15
Sphaerocystis	10	Oocystis	15
Total	94	Total	98

Appendix

Date	Station 2	#/ml	Station 3
21 July	Cyclotella	15,795	Cyclotella
	Anabaena	1,009	Anabaena
	Glenodinium	230	Glenodinium
	Carteria	225	Carteria
	Total	17,504	Total
31 July	Cyclotella	4,411	Anabaena
	Anabaena	2,027	Cyclotella
	Glenodinium	157	Glenodinium
	Carteria	115	Carteria
	Total	6,891	Total
19 Aug	Cyclotella	7,613	Cyclotella
	Glenodinium	1,158	Anabaena
	Anabaena	1,105	Navicula
	Franceia	371	Carteria
	Total	11,467	Total
28 Aug	Cyclotella	5,021	Cyclotella
	Anabaena	606	Anabaena
	Glenodinium	328	Navicula
	Navicula	207	Glenodinium
	Total	6,366	Total
17 Sept	Cyclotella	2,585	Anabaena
	Anabaena	1,236	Cyclotella
	Glenodinium	400	Navicula
	Navicula	118	Glenodinium
	Total	4,710	Total

II. (continued)

#/ml	Station 4	#/ml	Station 6	#/ml
1,361				
510				
57				
54				
2,160				
1,999	Anabaena	118	Anabaena	193
1,087	Navicula	74	Cyclotella	118
31	Cyclotella	39	Navicula	116
30	Glenodinium	25	Oscillatoria	36
3,260	Total	366	Total	510
748	Anabaena	396	Anabaena	1,622
739	Navicula	297	Navicula	315
223	Cyclotella	236	Cyclotella	105
94	Glenodinium	84	Oscillatoria	39
2,045	Total	1,169	Total	2,463
2,278	Anabaena	2,318	Anabaena	1,283
1,179	Cyclotella	230	Navicula	361
177	Navicula	210	Cyclotella	98
113	Oscillatoria	46	Carteria	34
4,589	Total	2,916	Total	1,860
2,253	Anabaena	2,062	Anabaena	2,232
246	Navicula	402	Navicula	362
162	Oscillatoria	51	Cyclotella	92
85	Cyclotella	46	Carteria	38
3,137	Total	2,900	Total	2,809

Appendix II

Date	Station 2	#/ml	Station 3
2 Oct.	Cyclotella	3,969	Anabaena
	Anabaena	1,407	Cyclotella
	Glenodinium	62	Navicula
	Tetracyclus	53	Carteria
	Total	5,783	Total
16 Oct.	Cyclotella	10,326	Cyclotella
	Tetracyclus	767	Anabaena
	Anabaena	564	Tetracyclus
	Glenodinium	499	Fragilaria
	Total	12,608	Total
30 Oct.	Cyclotella	17,197	Cyclotella
	Chamydomonas	666	Anabaena
	Carteria	335	Tetracyclus
	Platymonas	305	Fragilaria
	Total	19,313	Total
20 Nov.	Cyclotella	3,854	Cyclotella
	Chlamydomonas	597	Anabaena
	Carteria	295	Carteria
	Anabaena	171	Chlamydomonas
	Total	5,215	Total
22 Dec.	Cyclotella	2,696	Cyclotella
	Carteria	197	Carteria
	Chlamydomonas	105	Oscillatoria
	Oscillatoria	20	Fragilaria
	Total	3,064	Total

. (continued)

#/ml	Station 4	#/ml	Station 6	#/ml
1,675			Anabaena	1,561
989			Navicula	115
88			Cyclotella	56
46			Carteria	48
3,011			Total	1,871
7,577	Anabaena	814	Anabaena	1,455
584	Cyclotella	766	Cyclotella	514
301	Navicula	85	Navicula	81
85	Carteria	61	Tetracyclus	19
8,925	Total	1,927	Total	2,157
8,140	Cyclotella	3,449	Cyclotella	2,902
150	Anabaena	510	Anabaena	449
126	Carteria	49	Carteria	43
90	Navicula	41	Tetracyclus	34
8,825	Total	4,198	Total	3,611
3,725	Cyclotella	3,670	Cyclotella	2,837
127	Anabaena	209	Anabaena	102
62	Franceia	56	Fragilaria	42
562	Carteria	49	Tetracyclus	42
4,199	Total	4,115	Total	3,105
2,685	Cyclotella	2,305	Cyclotella	2,590
66	Carteria	43	Oscillatoria	28
10	Oscillatoria	13	Carteria	23
4	Navicula	7	Navicula	13
2,802	Total	2,358	Total	2,667

Appendix II.

Date	Station 2	#/ml	Station 3
29 Jan	Chlamydomonas	72	Cyclotella
	Carteria	59	Carteria
	Glenodinium	59	Chlamydomonas
	Cyclotella	53	Stephanodiscus
	Total	388	Total
19 Feb	Stephanodiscus	53	Cyclotella
	Cyclotella	26	Stephanodiscus
	Oocystis	20	Oocystis
	Navicula	16	Chlamydomonas
	Total	194	Total
29 Mar.	Cyclotella	1,079	Eudorina
	Eudorina	233	Cyclotella
	Fragilaria	66	Fragilaria
	Navicula	39	Stephanodiscus
	Total	1,600	Total

(continued)

#/ml	Station 4	#/ml	Station 6	#/ml
116	Cyclotella	92	Cyclotella	128
46	Carteria	33	Chlamydomonas	49
36	Stephanodiscus	26	Carteria	40
26	Chlamydomonas	16	Stephanodiscus	10
278	Total	190	Total	244
102	Cyclotella	293	Stephanodiscus	157
39	Stephanodiscus	214	Cyclotella	92
33	Chlamydomonas	36	Carteria	62
20	Carteria	33	Chlamydomonas	30
221	Total	607	Total	377
84				
67				
46				
20				
279				

Appendix II. (continued)

Depth (m)	Station 5		Station 5		Station 5	
	20 Aug	#/ml	28 Aug	#/ml	2 Oct	#/ml
0	Anabaena	537			Anabaena	469
	Cyclotella	364			Navicula	112
	Navicula	294			Cyclotella	79
	Oscillatoria	34			Phacotus	21
	Total	1,317			Total	723
5	Cyclotella	654	Anabaena	1,415	Anabaena	177
	Anabaena	146	Navicula	334	Navicula	159
	Navicula	173	Cyclotella	156	Cyclotella	92
	Oscillatoria	49	Fragilaria	33	Oscillatoria	23
	Total	1,281	Total	1,984	Total	508
10	Anabaena	540	Anabaena	436	Anabaena	270
	Cyclotella	390	Navicula	213	Cyclotella	207
	Navicula	224	Cyclotella	192	Navicula	115
	Fragilaria	38	Carteria	46	Carteria	36
	Total	1,394	Total	1,584	Total	666
15	Cyclotella	379	Anabaena	312	Anabaena	198
	Anabaena	161	Navicula	168	Cyclotella	105
	Navicula	139	Cyclotella	105	Navicula	39
	Fragilaria	31	Carteria	30	Carteria	10
	Total	1,054	Total	692	Total	623
20	Navicula	103	Cyclotella	103	Navicula	75
	Anabaena	16	Navicula	66	Cyclotella	40
	Carteria	15	Anabaena	47	Carteria	13
	Cyclotella	13	Fragilaria	18	Phacotus	13
	Total	333	Total	290	Total	156
30	Navicula	45	Navicula	48	Navicula	29
	Carteria	15	Cyclotella	33	Cyclotella	17
	Fragilaria	10	Anabaena	17	Phacotus	8
	Oscillatoria	7	Fragilaria	16	Glenodinium	3
	Total	98	Total	156	Total	66
40	Navicula	48	Navicula	35	Navicula	51
	Cyclotella	39	Cyclotella	26	Fragillaria	10
	Fragilaria	26	Anabaena	13	Anabaena	7
	Asterionella	7	Phacotus	10	Carteria	7
	Total	151	Total	125	Total	106

Appendix II. (continued)

Depth (m)	Station 5		Station 5		Station 5	
	9 May	#/ml	17 June	#/ml	18 July	#/ml
0	Fragilaris	59	Fragilaria	20	Cyclotella	126
	Oocystis	56	Sphaerocystis	7	Anabaena	102
	Asterionella	15	Oocystis	5	Phacotus	12
	Gomphonema	8	Carteria	3	Carteria	8
	Total	162	Total	53	Total	296
5	Fragilaria	53	Sphaerocystis	36	Anabaena	169
	Oocystis	34	Fragilaria	20	Cyclotella	143
	Asterionells	16	Oocystis	12	Oocystis	18
	Sphaerocystis	13	Ceratium	2	Franceia	13
	Total	157	Total	71	Total	404
10	Oocystis	56	Sphaerocystis	41	Cyclotella	46
	Fraqilaris	41	Oocystis	8	Franceia	26
	Sphaerocystis	15	Ceratium	8	Anabaena	25
	Cyclotella	10	Fragilaria	5	Carteria	20
	Total	171	Total	80	Total	228
15	Fraqilaria	43	Sphaerocystis	16	Cyclotella	43
	Oocystis	26	Fragilaria	9	Anabaena	31
	Sphaerocystis	12	Oocystis	5	Oocystis	28
	Asterionella	8	Chlorella	3	Fragilaria	10
	Total	115	Total	43	Total	156
20	Fraqilaris	30	Fragilaria	12	Fraqilaria	54
	Oocystis	30	Sphaerocystis	10	Oocystis	31
	Sphaerocystis	12	Cyclotella	8	Pandorina	8
	Asterionella	5	Oocystis	3	Phacotus	7
	Total	90	Total	40	Total	144
30	Oocystis	26	Fragilaria	10	Fragilaria	23
	Fraqilaria	10	Oocystis	8	Oocystis	18
	Cyclotella	7	Glenodinium	3	Cymbella	10
	Sphaerocystis	3	Hyalotheca	3	Cyclotella	8
	Total	64	Total	31	Total	77
40	Oocystis	16	Oocystis	10	Fragilaria	20
	Cyclotella	10	Fragilaria	7	Cyclotella	12
	Stephanodiscus	8	Navicula	3	Oocystis	11
	Fragilaria	5	Cyclotella	2	Navicula	2
	Total	51	Total	25	Total	61

Appendix II. (continued)

Depth (m)	Station 5		Station 5		Station 5	
	30 Oct	#/ml	20 Nov	#/ml	23 Dec	#/ml
0	Cyclotella	1,919	Cyclotella	2,892	Cyclotella	2,534
	Anabaena	454	Anabaena	131	Carteria	30
	Navicula	50	Franceia	69	Anabaena	13
	Carteria	36	Carteria	56	Navicula	13
	Total	2,604	Total	3,260	Total	2,598
5	Cyclotella	1,646	Cyclotella	2,906	Cyclotella	2,458
	Anabaena	141	Anabaena	72	Carteria	26
	Navicula	43	Fragilaria	33	Anabaena	10
	Fragilaria	39	Carteria	23	Fragilaria	3
	Total	1,935	Total	3,091	Total	2,514
10	Cyclotella	2,148	Cyclotella	3,114	Cyclotella	2,060
	Anabaena	788	Anabaena	138	Carteria	67
	Navicula	63	Franceia	66	Chlamydomonas	8
	Fragilaria	53	Melosira	33	Fragilaria	3
	Total	3,164	Total	3,459	Total	2,150
15	Cyclotella	1,983	Cyclotella	3,001	Cyclotella	2,164
	Anabaena	299	Anabaena	69	Carteria	36
	Fragilaria	36	Fragilaria	33	Anabaena	33
	Navicula	33	Carteria	33	Navicula	18
	Total	2,440	Total	3,203	Total	2,294
20	Cyclotella	1,729	Cyclotella	3,619	Cyclotella	3,090
	Anabaena	851	Anabaena	76	Anabaena	36
	Navicula	52	Carteria	43	Carteria	33
	Carteria	36	Fragilaria	33	Navicula	16
	Total	2,755	Total	3,849	Total	3,191
30	Cyclotella	139	Cyclotella	3,236	Cyclotella	2,209
	Navicula	59	Anabaena	134	Carteria	30
	Carteria	10	Carteria	59	Anabaena	21
	Phacotus	10	Fragilaria	33	Navicula	8
	Total	269	Total	3,539	Total	2,292
40	Cyclotella	69	Cyclotella	1,520	Cyclotella	2,664
	Navicula	43	Anabaena	29	Anabaena	25
	Carteria	13	Navicula	29	Navicula	20
	Achnanthe	7	Melosira	25	Total	20
	Total	141	Total	1,637	Total	2,768