Absence of Long-term Changes in the Microbiology of Freshwater Ponds Treated With Matacil®

D. J. Wildish and J. K. Elner

Biological Station,
St. Andrews, N.B., E0G 2X0

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ABSTRACT

Wildish, D. J., and J. K. Elner. 1981. Absence of long-term changes in the microbiology of freshwater ponds treated with Matacil®. Can. Tech. Rep. Fish. Aquat. Sci. 1049: iii + 17 p.

Spraying with Matacil® at up to 700 g ha^{-1} in an attempt to simulate contamination caused by aerial forest spraying for control of the spruce budworm had no immediate effect (1 d) on algal primary production, species composition of microalgae and invertebrates, numbers and biomass of heterotrophic bacteria, nor upon dissolved organic carbon and oxygen concentrations. No indications of Matacil®-caused effects on pond microbiology were found during long-term observations which continued for 1 yr following spraying.

Key words: Freshwater ponds, microbial ecology, forest pesticide side-effects

RÉSUMÉ

Wildish, D. J., and J. K. Elner. 1981. Absence of long-term changes in the microbiology of freshwater ponds treated with Matacil®. Can. Tech. Rep. Fish. Aquat. Sci. 1049: iii + 17 p.

L'arrosage au Matacil® à des concentrations allant jusqu'à 700 g ha^{-1} dans le but de simuler la contamination résultant de l'arrosage aérien des forêts dans le contrôle de la tordeuse des bourgeons de l'épinette n'eut pas d'effet immédiat (après 1 jour) sur la production primaire d'algues, la composition par espèce des micro-algues et des invertébrés, le nombre et la biomasse des bactéries hétérotrophes, non plus que sur les concentrations de carbone organique et d'oxygène dissous. Nous n'avons observé aucun effet du pesticide sur la microbiologie des étangs au cours d'observations à long term effectuées dans l'année qui suivit l'arrosage.

INTRODUCTION

The specific objectives of this work were to assess changes in the functioning of major physiological groups of freshwater micro-organisms and to determine the chemical fate of the active ingredient of Matacil®, the carbamate aminocarb, following simulated aerial spraying.

Matacil® was used in 1978 and 1979 to control spruce budworm in New Brunswick forests. Further details of the rationale for this study are given in a previous report (Elner and Wildish 1981).

Presented here are the short-term observations made on the day of spraying in May/June 1980, some chemical analyses of pond water taken soon after spraying to determine aminocarb metabolites, and a continuation of previously reported long-term observations on pond microbiology up to September 1980 (Elner and Wildish 1981) for a further 8-mo period to June 1981, after which the field experiment was terminated.

METHODS

LONG-TERM OBSERVATIONS

Sampling locations, techniques and analysis were carried out as previously described in Elner and Wildish (1981).

SHORT-TERM OBSERVATIONS

Sampling locations, simulated aerial spray methodology, sampling techniques and analyses were as used previously (Elner and Wildish 1981) except that the frequency of sampling was increased to 10 samples/day.

CHEMICAL ANALYSES

A few water samples from the ponds were analyzed for aminocarb metabolites by the method of Brun and MacDonald (1978). The final extract was dissolved in acetonitrile for HPLC. A reverse phase Lichrosorb RP-2 column (250 x 4.6 mm internal diameter) was used in a Varian 5000 LC with a fixed wavelength UV detector at 254 nm. The mobile phase consisted of 20% acetonitrile in water at a flow rate of 2 mL/min. Authentic metabolite standards were used to help identify unknown peaks.

Water samples of 1-L volume were obtained from the ponds in brown glass bottles and extracted in the field with 15 mL pesticide-grade ethyl acetate. Bottles were stored at 4°C until final extraction and analyses in the Research and Productivity Council laboratories, Fredericton, N.B.

RESULTS AND DISCUSSION

LONG-TERM OBSERVATIONS

Records of temperature, Eh and pH are shown in Appendix 1. Dissolved oxygen and dissolved organic matter concentrations are recorded in Appendices 2 and 3. The plant pigment record for the study (Appendices 4-7) indicates high levels of chlorophyll a during January and February when the ponds were ice-covered. As in the winter 1979-80 (Elner and Wildish 1981), the following winter phytoplankton maximum was also composed of a multispecies community. The 1980-81 winter differed from the previous one in that there was an extensive period of snow-cover beginning in December 1980. The phytoplankton maximum coincided with zero levels of dissolved oxygen in January (Appendix 2). Presumably the bloom was in a stationary/senescent stage when sampled by us and heterotrophic bacterial activity had utilized all available oxygen in the temporarily closed system. Dissolved oxygen levels would not increase again until a new bloom had occurred or after ice-out.

The major groups of microalgae and invertebrates found during this study of pond water are listed in Appendices 8-11.

Pond water ATP Levels (Appendix 12) and primary production (Appendix 13) showed no marked systematic differences. Pond B had the highest production level in January 1981 during the winter bloom.

SHORT-TERM OBSERVATIONS

The morphometry of the four ponds and the area of associated marshland are described in Elner and Wildish (1981). Matacil® was sprayed onto the ponds in a fine mist from a Solo 423 knapsack sprayer (Boynton and Smith 1971) in three treatments (Table 1).

Table 1. Matacil® treatment dosage and sampling date.

Pond	Matacil® treatment (g aminocarb ha^{-1})	Application and sampling date
A	0-control	03.06.80
B	140	27.05.80
C	700	03.05.80
D	280	29.05.80

The microbiological and physiochemical observations made in a 14-h period after spraying are presented as graphs (Fig. 1-10) or tables (Appendices 14-17). Chemical analyses for aminocarb in water samples taken during this study are presented elsewhere (Elner et al. 1981) and indicate that a maximum subsurface concentration of 0.055 mg/L is reached.

Maximum temperature fluctuations occurred on 2 d that were dry and sunny (Ponds B and D - Fig. 2). Temperatures on these 2 d ranged from 13 to 21°C. The dissolved oxygen curves for three of the ponds (Ponds A, B, and C - Fig. 4) followed a unimodal form as found by Odum (1956) and Lingeman and Ruardij (1981). Dissolved oxygen levels in Pond D showed a bimodal diel pattern as found less commonly by Edwards and Owens (1962), Kalbe (1972), and Lingeman and Ruardij (1981). The bimodal diel pattern of dissolved oxygen concentration was found to occur most frequently in May and June and to result from either persistently high irradiation levels at mid-day or a high rate of temperature

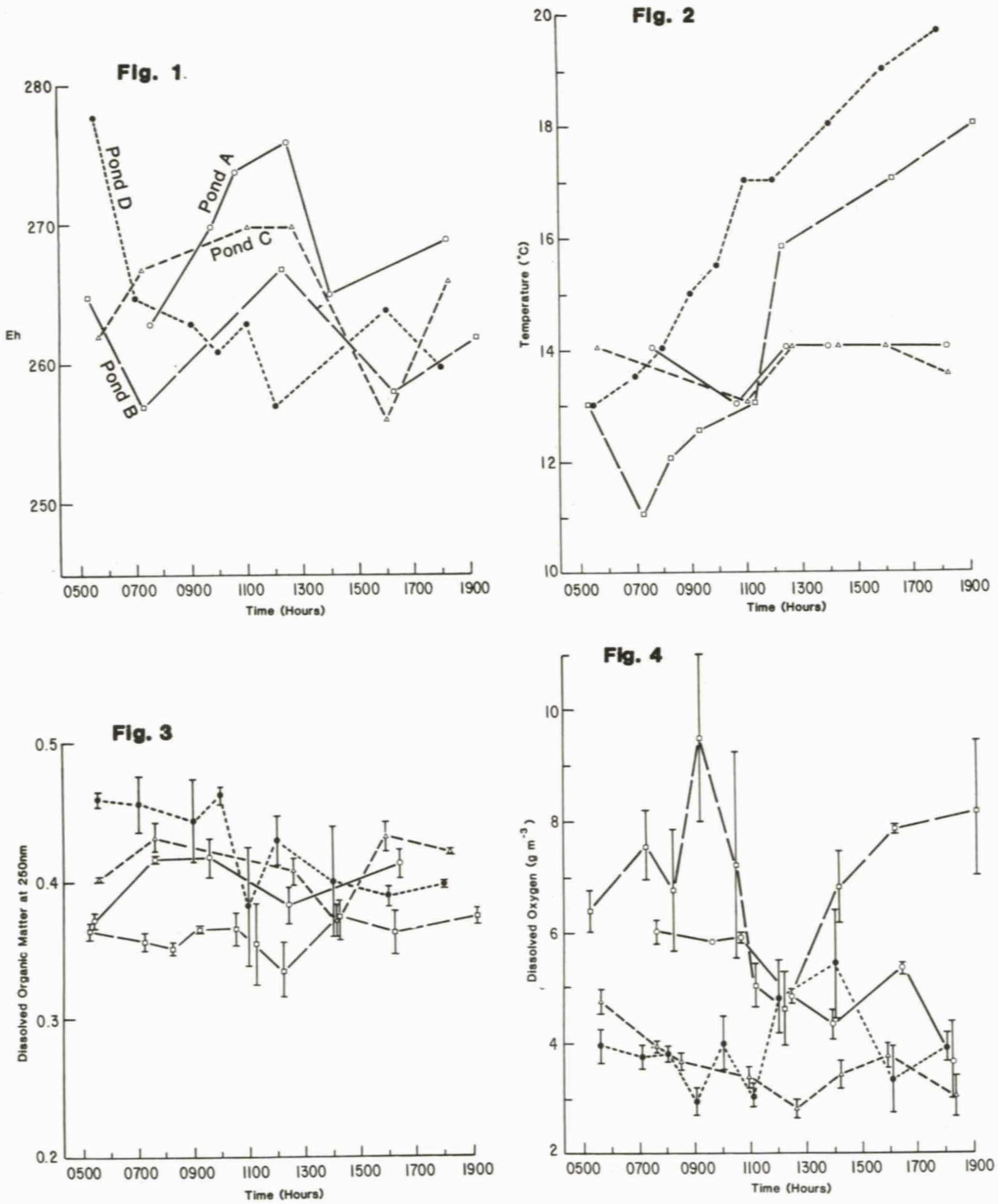


Fig. 1-4. Short-term changes in Eh (Fig. 1); temperature (Fig. 2); dissolved organic matter (Fig. 3); and dissolved oxygen concentration (Fig. 4). ○- Pond A, □- Pond B, △ - Pond C, ● - Pond D.

increase associated with this (Lingeman and Ruardij 1981). Dissolved oxygen saturation curves often reflect diel fluctuations in primary production as indicated by chlorophyll a levels. In our work, only in Ponds A and C does the dissolved oxygen follow the chlorophyll a concentration, with a 2- to 4-h lag period.

In all four ponds there were peaks in phytoplankton abundance in surface water during the morning and evening of the day of sampling, with an afternoon depression, the time of which varied between ponds (Appendices 14-17). Changes in phytoplankton abundance were followed by changes in chlorophyll a concentration (Fig. 5, 6, 7, and 8). The two major algal taxa involved were flagellate chlorophyta and diatoms (Appendices 14-17). Diel changes in the position of phytoplankton within the water column likely result from a combination of factors. These include:

- passive: e.g., sinking rates are increased by high temperature (Riley et al. 1949); turbulence which forces cells to less turbulent layers (Ganf and Horne 1975); and nutrient depletion (Ohle 1961).

- active: Flagellates are able to swim away from excessive radiation, whilst blue-green algae can move into or out of the surface water by formation and collapse of a gas vacuole (Fogg and Walsby 1971). Inactivation of the photosynthetic apparatus may occur around noon due to the high light intensity (Yentsch and Rhyther 1957; Kairesalo 1980).

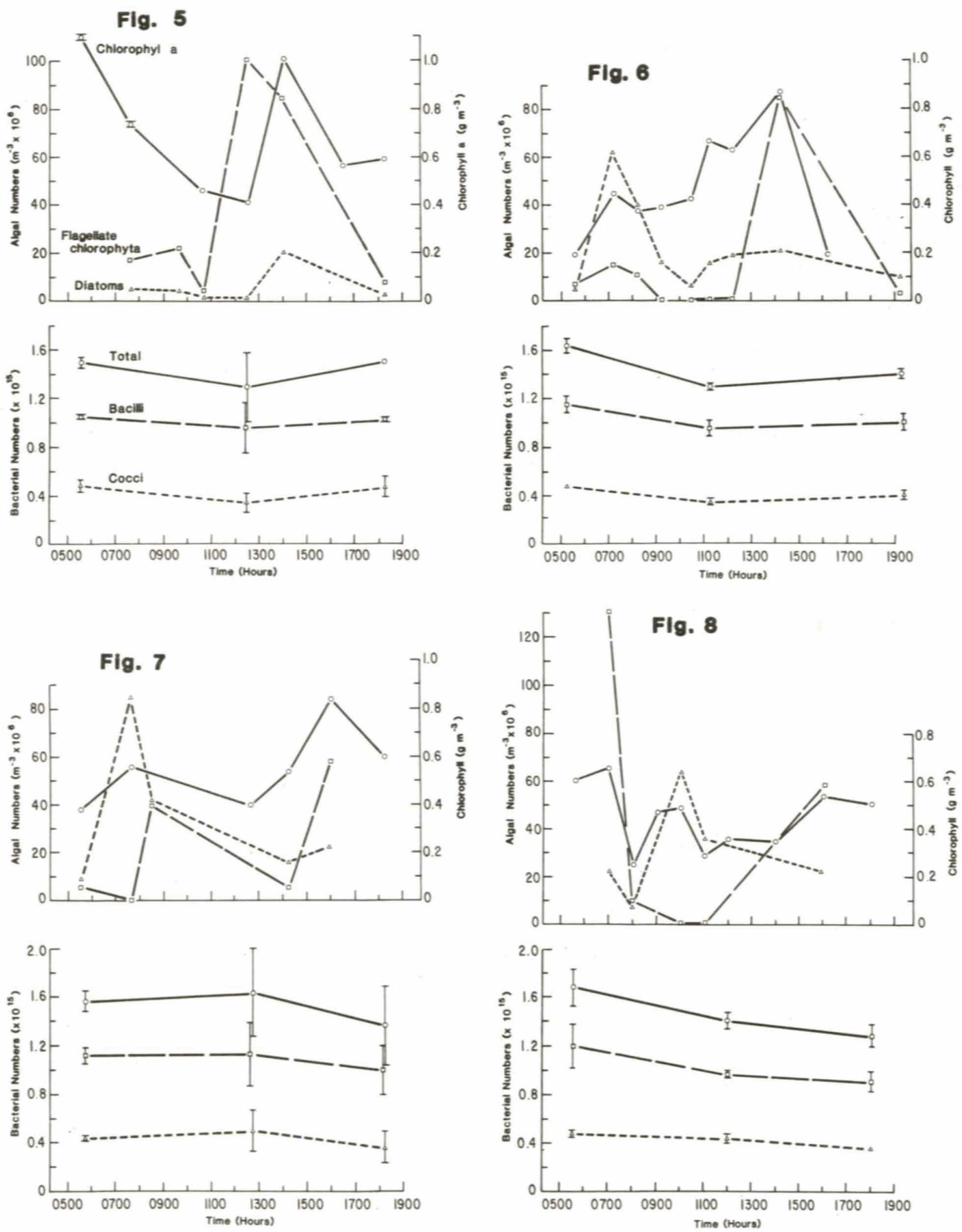
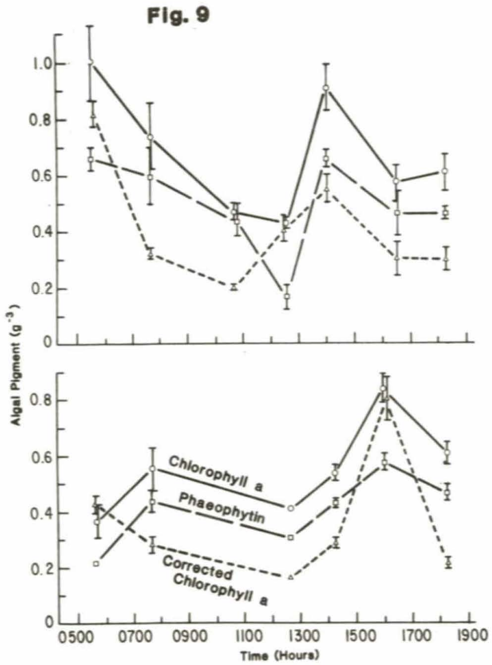


Fig. 5-8. Short-term changes in chlorophyll a, algal and bacterial numbers in each pond. Pond A (Fig. 5); Pond B (Fig. 6); Pond C (Fig. 7); and Pond D (Fig. 8). Refer to Fig. 5 for explanation of symbols.

It is these passive and/or active movements which result in the fluctuations recorded in chlorophyll a and dissolved oxygen concentrations. A detailed analysis of photosynthetic pigments (Fig. 9, 10) also supports the idea of diel changes in phytoplankton communities.

CHEMICAL ANALYSIS OF WATER FOR METABOLITES

The active ingredient of the formulated pesticide Matacil®, is aminocarb (see Fig. 11, Compound I). Aminocarb is known to undergo metabolism in plants and animals, to be biodegraded by microbes, and to undergo photodecomposition.



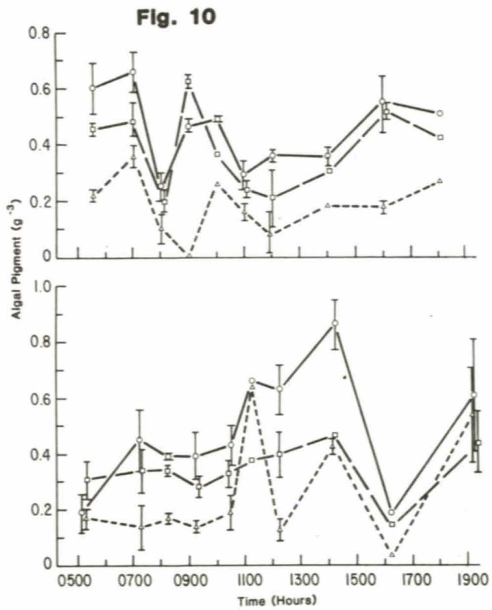


Fig. 9-10. Short-term variation in algal pigments. Fig. 9: Pond A - upper, Pond B - lower. Fig. 10: Pond C - upper, Pond D - lower. ○ - chlorophyll a, □ - phaeophytin, △ - chlorophyll a corrected for the presence of phaeophytin.

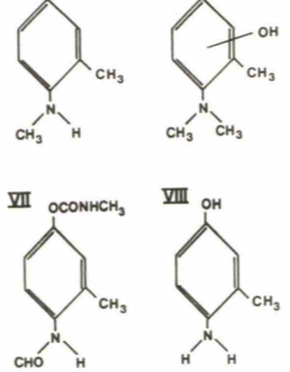


Fig. 11. 4-dimethylamino-3-methylphenyl N-methylcarbamate (Compound I, aminocarb) and its known metabolites identified by roman numerals in the text.

Metabolism in plants and animals involves a series of oxidation and demethylation steps producing numerous products, most of which are short-lived and some of which are unknown. The major products recognized by Kuhr and Dorough (1976) are:

- II 4-amino-3-methyl-phenyl N-methylcarbamate
- III 4-dimethylamino-3-methylphenyl N-(hydroxymethyl)carbamate
- IV 4-(methylamino)-3-methylphenyl N-methylcarbamate

Compounds I and III have equal anticholinesterase activity, whereas II and IV have ten times more activity (Kuhr and Dorough 1976). In plants, further metabolism causes inactivation by conjugation of the products with tannin and plant wall substances.

Microbial biodegradation studies (see Steeves 1971) show that, in addition to the above, microbes can also ring hydroxylate carbamates (Fig. 11) as well as cleave them to the corresponding phenol, thus producing the following additional products:

- V ring hydroxylation products of uncertain structure
- VI 4-dimethylamino-3-methylphenol

x

These findings have been confirmed by Balba et al. (1974), who found products V and VI following the use of the ascorbic acid oxidation system which is designed to chemically mimic biodegradation pathways.

Photodecomposition occurs under certain conditions (Addison et al. 1974), the major product being Compound V.

Nine metabolites were described by Brun and MacDonald (1978) from freshwater of which only three were found in our samples (Table 2). The structure of these metabolites is shown in Fig. 11 and includes also:

- VII 4-(formylamino)-3-methylphenyl N-methylcarbamate
- VIII 4-amino-3-methylphenol

The appearance of Compound VII in the control pond casts some doubt on the reliability of the analyses.

Table 2. Concentration of aminocarb metabolites in control and treatment ponds.

Pond	Time after spray (d)	Metabolite concentration (ppb) VI	VII	VIII
A	10	0	1.2	0
C	3	10.4	0	0
	10	0	0	0
	16	0	1.4	0
	24	0	3.2	0
D	6	3.3	0	12.1
	13	0	0	0
	19	0	0	0
	27	0	0	0
B	8	4.5	0	13.2
	15	0	0	0
	21	0	1.7	0
	29	0	0	0

CONCLUSIONS

In chemical accountability studies with Matacil® (Elner et al. 1981), it was shown that a maximum of 55 µg/L of aminocarb would be found in lentic subsurface water following a simulated aerial application of 140 g ha^{-1}, and that this peak would not be reached until 22- to 72-h post-spray. Even at 5x this application rate, 700 g ha^{-1}, the subsurface concentration would be only 117 µg/L at 24 h (Elner et al. 1981). Primary production by the natural algal communities of the experimental ponds was not affected by ethanol solubilized Matacil R at concentrations below 450 µg/L (Elner et al. 198[illegible]) and, even at concentrations above this, where there was a marked reduction of carbon assimilation, production rapidly recovered within 72 h due to the existence of resistant algal cells.

If this experiment were repeated with another pesticide, it would obviously be better to extend the observations into the second and third days following spraying. However, we do not think, even if this were done, that it would lead to a change in our conclusion that spraying Matacil® at up to 5x the operational spray dosage rate had any immediate effect on algal primary production, or species composition of microalgae and invertebrates, hetetrophic bacterial numbers and biomass, dissolved organic carbon, and dissolved oxygen levels. Previous authors such as Weinberger and Rea (1981) found an increase in the lag phase of a laboratory culture of Chlorella at concentrations greater than 25 µg/L of the surfactant nonyphenol, which is used in the Matacil® formulation. Matacil® contains 2x as much nonylphenol by weight as it does aminocarb and it is possible that the reported toxic effects are due to nonylphenol. Our field observations of primary production show that these laboratory observed effects are of little consequence. More serious effects noted by Weinberger and Rea (1981), inclusive of flagellae distortion and architectural disruption of cell ultrastructure, occurred at levels of >500 µg/L nonylphenol. This concentration is much greater than expected field levels of nonylphenol. Moreover, the Chlorella culture was able to recover rapidly.

We have now followed microbiological activity over a complete 12-mo period following spraying and we have found no indications of a long-term effect. This is consistent with the known rapid degradation of aminocarb in pond water, a degradation rate constant of 69-205/yr being found within 95% confidence limits (Elner et al. 1981). Because a suitable technique for determining aminocarb in organic sediments was not available, we were not able to determine concentrations present there. Sediments could act as a sink for aminocarb and both the parent compound and its microbially produced metabolites might persist there for a longer period than in water.

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Appendix 1. Temperature, pH and Eh from October 1980-June 1981 in the study ponds (temperature - °C; pH - pH units, Eh - relative to the normal hydrogen electrode).

Date	Pond A			Pond B			Pond C			Pond D		
	Temp.	pH	Eh	Temp.	pH	Eh	Temp.	pH	Eh	Temp.	pH	Eh
21.10.80	5	6.1		5	6.0		5	6.1		5	5.7	
09.12.80	0			0			0			0		
19.01.81	0	6.4		0	5.9		0	6.5		0	6.0	
26.01.81	0	6.6		0	6.4		0	6.3		0	5.9	
02.03.81	0	6.1		0	6.1		0	6.0		0	5.7	
02.06.81	11	6.2	257	18	4.9	261	12	6.2	260	15	5.6	263

Appendix 2. Dissolved oxygen concentrations from October 1980-June 1981 in the study ponds.

Date	Dissolved oxygen ($g\ m^3$) $\pm$ S.E.			
	Pond A	Pond B	Pond C	Pond D
21.10.80	3.60 $\pm$ 0.10	4.06 $\pm$ 0.14	5.62 $\pm$ 0.25	5.82 $\pm$ 0.20
09.12.80	8.99 $\pm$ 0.08	10.83 $\pm$ 0.00	7.73 $\pm$ 0.03	2.16 $\pm$ 0.05
19.01.81	3.16 $\pm$ 0.25	0.00 $\pm$ 0.00	0.00 $\pm$ 0.00	0.00 $\pm$ 0.00
26.01.81	4.44 $\pm$ 0.22	0.00 $\pm$ 0.00	8.31 $\pm$ 0.13	0.00 $\pm$ 0.00
02.03.81	7.14 $\pm$ 0.01	8.14 $\pm$ 0.69	9.66 $\pm$ 0.11	2.41 $\pm$ 0.01
02.06.81	4.91 $\pm$ 0.06	5.24 $\pm$ 0.16	6.11 $\pm$ 0.11	6.54 $\pm$ 0.25

Appendix 3. Dissolved organic matter as arbitrary absorbance units at 250 nm from October 1980-June 1981 in the study ponds.

Date	Dissolved organic matter as measured at 250 nm $\pm$ S.E.			
	Pond A	Pond B	Pond C	Pond D
21.10.80	0.56 $\pm$ 0.007	0.29 $\pm$ 0.010	0.52 $\pm$ 0.007	0.49 $\pm$ 0.105
09.12.80	0.27 $\pm$ 0.002	0.18 $\pm$ 0.006	0.32 $\pm$ 0.001	0.32 $\pm$ 0.001
19.01.81	0.53 $\pm$ 0.004	1.51 $\pm$ 0.129	0.37 $\pm$ 0.001	2.34 $\pm$ 0.003
26.01.81	0.43 $\pm$ 0.012	2.30 $\pm$ 0.030	0.36 $\pm$ 0.002	1.85 $\pm$ 0.022
02.03.81	0.39 $\pm$ 0.002	0.33 $\pm$ 0.003	0.31 $\pm$ 0.010	0.61 $\pm$ 0.012
02.06.81	0.50 $\pm$ 0.014	0.31 $\pm$ 0.006	0.61 $\pm$ 0.035	0.46 $\pm$ 0.012

Appendix 4. Chlorophyll a from October 1980-June 1981 in the study ponds (determined by Parsons and Strickland (1963) method).

Date	Chlorophyll a (g m^3) ± S.E. Pond A	Pond B	Pond C	Pond D
21.10.80	1.174 ± 0.185	0.192 ± 0.030	0.138 ± 0.016	0.418 ± 0.084
09.12.80	0.224 ± 0.012	0.129 ± 0.024	0.138 ± 0.010	0.242 ± 0.015
19.01.81	1.656 ± 0.136	10.783 ± 1.233	0.392 ± 0.118	7.450 ± 0.560
26.01.81	0.392 ± 0.018	3.520 ± 0.112	0.294 ± 0.118	3.276 ± 0.711
02.03.81	0.182 ± 0.012	0.221 ± 0.003	0.196 ± 0.010	0.262 ± 0.024
02.06.81	0.456 ± 0.029	0.526 ± 0.079	0.253 ± 0.018	1.227 ± 0.104

Appendix 5. Chlorophyll a after correction for phaeophytins from October 1980-June 1981 in the study ponds (determined by the technique of Moss 1967).

Date	Chlorophyll a (g m^3) ± S.E. Pond A	Pond B	Pond C	Pond D
21.10.80	0.833 ± 0.030	0.099 ± 0.031	0.074 ± 0.009	0.245 ± 0.034
09.12.80	0.122 ± 0.007	0.085 ± 0.014	0.084 ± 0.003	0.153 ± 0.016
19.01.81	0.903 ± 0.075	5.015 ± 1.145	0.288 ± 0.072	2.560 ± 0.390
26.01.81	0.203 ± 0.096	1.708 ± 0.117	0.186 ± 0.066	1.157 ± 0.645
02.03.81	0.050 ± 0.004	0.109 ± 0.008	0.116 ± 0.002	0.175 ± 0.049
02.06.81	0.213 ± 0.076	0.156 ± 0.018	0.129 ± 0.021	0.594 ± 0.048

Appendix 6. Phaeophytin from October 1980-June 1981 in the study ponds.

Date	Phaeophytin (g m^3) ± S.E. Pond A	Pond B	Pond C	Pond D
21.10.80	0.671 ± 0.225	0.170 ± 0.050	0.110 ± 0.015	0.255 ± 0.062
09.12.80	0.168 ± 0.008	0.075 ± 0.007	0.076 ± 0.005	0.155 ± 0.011
19.01.81	0.547 ± 0.236	5.780 ± 0.710	0.260 ± 0.065	1.768 ± 1.332
26.01.81	0.196 ± 0.094	2.132 ± 0.065	0.196 ± 0.081	2.304 ± 0.473
02.03.81	0.143 ± 0.006	0.122 ± 0.001	0.133 ± 0.007	0.255 ± 0.075
02.06.81	0.378 ± 0.025	0.451 ± 0.016	0.175 ± 0.015	1.065 ± 0.236

Appendix 7. Changes in carotenoids from October 1980-June 1981 in the study ponds.

Date	Carotenoids (g m^3) ± S.E. Pond A	Pond B	Pond C	Pond D
21.10.80	0.688 ± 0.127	0.194 ± 0.055	0.105 ± 0.017	0.440 ± 0.126
09.12.80	0.137 ± 0.012	0.136 ± 0.013	0.077 ± 0.002	0.233 ± 0.006
19.01.81	1.528 ± 0.422	5.222 ± 0.778	0.422 ± 0.147	3.893 ± 0.037
26.01.81	0.000 ± 0.024	0.454 ± 0.006	0.104 ± 0.032	2.522 ± 0.195
02.03.81	0.016 ± 0.003	0.035 ± 0.001	0.036 ± 0.006	0.067 ± 0.003
02.06.81	0.286 ± 0.237	0.606 ± 0.040	0.151 ± 0.026	1.758 ± 0.205

Appendix 8. Major taxa of microalgae and invertebrates from Pond A (organism x 10^6 m^{-3})

Taxa \ Date	21.10.80	19.01.81	26.01.81	02.03.81	02.06.81
Total chlorophyceae	38.75	205.00	5.00	116.25	33.75
Filamentous	10.00	10.00			1.25
Flagellate	28.75	15.00		70.00	12.50
Bulbochaete					
Chloroccus		145.00		5.00	21.25
Chlamydomonas			5.00	41.25	
Scenedesmus		35.00			
Pediastrum					
Total diatoms	17.50	390.00	12.50	56.25	5.00
Total centrales			2.50	6.25	
Total pennales	17.50	390.00	10.00	50.00	5.00
Coscinodiscus subtilis				2.50	
Cyclotella			2.50	3.75	
Melosira distans					
Achnanthes					
Amphora					
Cocconeis		5.00			
Cymbella					
Diatoma					
Eunotia	3.75	55.00		2.50	
Frustulia vulgaris		15.00			
Gomphonema	1.25	20.00		13.75	
G. acuminatum		20.00			
Meridion circulare		.10.00		21.25	
Navicula	8.75	55.00			
Nitzchia		15.00			
Pinnularia		15.00	3.75		
P. major		25.00			
Synedra		15.00			
Tabellaria flocculosa (long type)		95.00		3.75	1.25
T. flocculosa	3.75		6.25	2.50	
Total desmids			5.00		
Closterium moniliforme					
Cosmarium reniforme					
Desmidium baileyi			5.00		
Euastrum didelta					
Pleurotaenium nodosum					
Sphaerozoma filiformis					
Staurastum inconspicuum					
S. subcruciatum					
Tetraedon constrictum					
Xanthidium ornatum					
X. sansibarense					
Total chrysophyceae		85.00			13.75
Dinobryon					7.50
Mallomonas		85.00			6.25
Total dinoflagellates					
Glenodinium					
Total blue greens	16.25	75.00	22.50		17.50
Filamentous	16.25	35.00			17.50
Anabaena		20.00	17.50		
Microcystis					
Oscillatoria		20.00	5.00		
Rotifer					

Appendix 9. Major taxa of microalgae and invertebrates from Pond B (organism x 10^6 m^{-3}).

Taxa \ Date	21.10.80	19.01.81	26.01.81	02.03.81	02.06.81
Total chlorophyceae	5.00	60.00			28.75
Filamentous			21.25		6.25
Flagellate	5.00		13.75		22.50
Bulbochaete					
Chloroccus		10.00	3.75		
Chlamydomonas					
Scenedesmus		35.00	5.00		
Pediastrum		15.00			
Total diatoms	3.75	1230.00	87.50	18.75	
Total centrales					
Total pennales	3.75	1230.00	87.50	18.75	
Coscinodiscus subtilis					
Cyclotella					
Melosira distans					
Achnanthes					
Amphora		10.00			
Cocconeis					
Cymbella					
Diatoma			6.25		
Eunotia	3.75	125.00	20.00		
Frustulia vulgaris		115.00	7.50		
Gomphonema		20.00			
G. acuminatum					
Meridion circulare				8.75	
Navicula		225.00	16.25		
Nitzchia					
Pinnularia		35.00			
P. major		5.00	1.25		
Synedra		10.00			
Tabellaria flocculosa (long type)		235.00	28.75	6.25	
T. flocculosa		275.00	7.50	3.75	
Total desmids		150.00			
Closterium moniliforme					
Cosmarium reniforme		26.00	1.25		13.75
Desmidium baileyi			2.50		
Euastrum didelta					
Pleurotaenium nodosum			6.25		
Sphaerozoma filiformis		30.00			
Staurastum inconspicuum					
S. subcruciatum		65.00	6.25		
Tetraedon constrictum					
Xanthidium ornatum					
X. sansibarense		30.00			
Total chrysophyceae					
Dinobryon					
Mallomonas			8.75		
Total dinoflagellates					
Glenodinium					
Total blue greens	8.75	210.00		3.75	
Filamentous	8.75			3.75	
Anabaena		10.00			
Microcystis		200.00	2.50		
Oscillatoria			3.75		
Rotifer					

Appendix 10. Major taxa of microalgae and invertebrates from Pond C (organisms x 10^6 m^{-3}).

Taxa \ Date	21.10.80	19.01.81	26.01.81	02.03.81	02.06.81
Total chlorophyceae	12.50	80.00	6.25		6.25
Filamentous	11.25	60.00			2.50
Flagellate	11.25		6.25		
Bulbochaete					
Chloroccus					3.75
Chlamydomonas					
Scenedesmus		20.00			
Pediastrum					
Total diatoms	13.75	330.00	12.50	45.06	
Total centrales	3.75	15.00		3.75	
Total pennales	11.25	315.00	12.50	41.25	
Coscinodiscus subtilis					
Cyclotella				3.75	
Melosira distans	3.75	15.00			
Achnanthes					
Amphora		20.00			
Cocconeis					
Cymbella				3.75	
Diatoma					
Eunotia		55.00			
Frustulia vulgaris		10.00		3.75	
Gomphonema		10.00	7.50		
G. acuminatum					
Meridion circulare					
Navicula	6.25	20.00	5.00	6.25	
Nitzchia					
Pinnularia				2.50	
P. major		10.00		6.25	
Synedra		15.00			
Tabellaria flocculosa (long type)		30.00		3.75	
T. flocculosa	2.50	15.00		2.50	
Total desmids					
Closterium moniliforme					
Cosmarium reniforme					
Desmidium baileyi					
Euastrum didelta					
Pleurotaenium nodosum					
Sphaerozoma filiformis					
Staurastum inconspicuu					
S. subcruciatum					
Tetraedon constrictum					
Xanthidium ornatum					
X. sansibarense					
Total chrysophyceae		10.00			3.75
Dinobryon					
Mallomonas		10.00			3.75
Total dinoflagellates					
Glenodinium					
Total blue greens	2.50	35.00	21.50		
Filamentous	2.50				
Anabaena					
Microcystis		15.00			
Oscillatoria		20.00	21.25		
Rotifer					3.75

Appendix 11. Major taxa of microalgae and invertebrates from Pond D (organisms x 10^6 m^{-3}).

Taxa \ Date	21.10.80	19.01.81	26.01.81	02.03.81	02.06.81
Total chlorophyceae	10.00	275.00	1.25	3.75	26.25
Filamentous	5.00	5.00	1.25		
Flagellate	5.00	60.00		3.75	17.50
Bulbochaete		15.00			
Chloroccus		150.00			8.75
Chlamydomonas					
Scenedesmus		35.00			
Pediastrum					
Total diatoms	13.15	620.00	37.50	21.25	
Total centrales					
Total pennales	13.75	620.00	37.50	21.25	
Coscinodiscus subtilis					
Cyclotella					
Melosira distans					
Achnanthes		5.00			
Amphora					
Cocconeis		15.00			
Cymbella					
Diatoma					
Eunotia	3.75	60.00	3.75	2.50	
Frustulia vulgaris		105.00	5.00	6.25	
Gomphonema					
G. acuminatum					
Meridion circulare		30.00			
Navicula		45.00			
Nitzchia	5.00				
Pinnularia		75.00			
P. major		35.00	3.75	6.25	
Synedra		45.00			
Tabellaria flocculosa (long type)		55.00			
T. flocculosa	1.25		1.25		
Total desmids		250.00		6.25	
Closterium moniliforme		25.00			
Cosmarium reniforme		20.00		2.50	
Desmidium baileyi				3.75	
Euastrum didelta					
Pleurotaenium nodosum					
Sphaerozoma filiformis					
Staurastum inconspicuu		130.00			
S. subcruciatum		25.00			
Tetraedon constrictum		20.00			
Xanthidium ornatum		5.00			
X. sansibarense		25.00			
Total chrysophyceae		10.00	87.50	3.75	
Dinobryon		10.00		3.75	
Mallomonas			87.50		
Total dinoflagellates		15.00			
Glenodinium		15.00			
Total blue greens	7.50	30.00	1.25		
Filamentous	7.50				
Anabaena					
Microcystis		5.00			
Oscillatoria		25.00	1.25		
Rotifer		10.00			

Appendix 12. ATP from October 1980-June 1981 in the study ponds.

Date	ATP ($mg\ m^{-3}$) ± S.E. Pond A	Pond B	Pond C	Pond D
20.01.80				0.124 ± 0.003
21.01.81	0.245 ± 0.021	0.174 ± 0.04	0.213 ± 0.05	0.170 ± 0.032
03.02.81	0.105 ± 0.014	0.170 ± 0.02	0.083 ± 0.019	0.181 ± 0.035

Appendix 13. Algal production measured by the ^{14}C assimilation method.

Date	Carbon assimilation ($\mu g\ Ch^{-1}\ m^{-3}$) Pond A	Pond B	Pond C	Pond D
06.08.80	21.32 ± 1.02			
16.09.80	35.11 ± 2.04			
19.01.81	2.33 ± 0.11	127.60 ± 4.25	1.36 ± 0.012	20.90 ± 0.001
26.01.81	3.72 ± 0.71	118.05 ± 4.25	0.92 ± 0.14	20.92 ± 0.26
02.06.81	15.45 ± 2.89	4.27 ± 0.82	6.99 ± 3.20	32.57 ± 3.22

Appendix 14. Major taxa of microalgae and invertebrates from Pond A (organism x 10^6 m^{-3}).

Taxa \ Time	0746	0950	1045	1230	1400	1815
Total chlorophyceae	22.5	37.5	3.7	113.7	91.7	8.7
Filamentous	3.7	10.0		3.7	3.7	2.5
Flagellate	16.2	22.5	3.7	102.6	85.5	6.2
Chloroccus						
Pandorina				5.0		
Scenedesmus	2.5	5.0		2.5	2.5	
Total diatoms	5.0	3.7	1.2	1.2	20.0	2.5
Total centrales						
Total pennales	5.0	3.7	1.2	1.2	20.0	2.5
Cyclotella						
Melosira distans						
Diatoma						
Eunotia					3.8	
Frustulia vulgaris						
Gomphonema						
Meridion circulare						
Navicula		1.2		1.2	12.5	2.5
Pinnularia						
Synedra						
Tabellaria flocculosa	2.5				2.5	
(long type)	1.2		1.2		1.2	
T. flocculosa						
Total desmids	5.0			5.0	2.5	5.0
Closterium moniliforme						
Cosmarium reniforme	5.0			5.0	2.5	5.0
Total chrysophyceae						
Dinobryon						
Total blue greens						
Filamentous						
Invertebrata						
Rotifers		2.5				
Ciliates		1.2				
Nematodes	48.7	291.2	118.7	10.0	13.7	37.5

Appendix 15. Major taxa of microalgae and invertebrates from Pond B (organism x 10^6 m^{-3}).

Taxa \ Time	0520	0715	0815	0915	1030	1115	1215	1415	1920
Total chlorophyceae	7.5	17.5	32.5	1.2	2.5	25.0	6.2	97.5	7.5
Filamentous	1.2		3.7			2.5	2.5	8.7	1.2
Flagellate	6.2	15.0	11.25			1.2	1.2	85.0	3.7
Chloroccus									
Pandorina									
Scenedesmus		2.5	17.5	1.2	2.5	21.2	2.5	3.7	2.5
Total diatoms	5.0	62.5	41.2	16.2	6.5	16.2	5.0	21.2	10.0
Total centrales			1.2	2.5					
Total pennales	5.0	62.5	40.0	13.7	6.5	16.2	3.7	21.2	5.0
Cyclotella			1.2	2.5					
Melosira distans									
Diatoma		17.5							
Eunotia		7.5	6.2	3.7		2.5	2.5		
Frustulia vulgaris									
Gomphonema		1.2	5.0		2.5				
Meridion circulare			3.7						
Navicula	1.2	7.5	5.0	5.0	3.7		2.5	11.2	1.2
Pinnularia		3.7	7.5			1.2			
Synedra									
Tabellaria flocculosa (long type)			5.0						
T. flocculosa	1.2	13.7	2.5	1.2		3.7	1.2	3.7	6.2
Total desmids									
Closterium moniliforme									
Cosmarium reniforme									
Total chrysophyceae						10.0		6.2	
Dinobryon						10.0		6.2	
Total blue greens	331.2	8.7	71.2	5.0		20.0	6.2	7.5	
Filamentous	331.2	8.7	71.2	5.0		20.0	6.2	7.5	
Invertebrata									
Rotifers				1.2					
Ciliates				1.2					
Nematodes									

Appendix 16. Major taxa of microalgae and invertebrates from Pond C (organism x 10^6 m^{-3}).

Taxa \ Time	0545	0750	0830	1415	1600
Total chlorophyceae	13.6	1.2	43.7	15.0	66.1
Filamentous	6.2	1.2	1.2		6.2
Flagellate	6.2		40.0	5.0	58.7
Chloroccus					
Pandorina					
Scenedesmus	1.2		2.5	10.0	1.2
Total diatoms	8.7	85.0	42.5	16.2	22.5
Total centrales	1.2	20.0	6.2		
Total pennales	7.5	65.0	36.2	16.2	22.5
Cyclotella					
Melosira distans	1.2	20.0	6.2		
Diatoma		5.0			
Eunotia		6.2	5.0		6.2
Frustulia vulgaris					
Gomphonema	1.2	3.7	1.2		
Meridion circulare					
Navicula	1.2	20.0		6.2	
Pinnularia			1.2		
Synedra			1.2	1.2	1.2
Tabellaria flocculosa (long type)		13.7			1.2
T. flocculosa			7.5	8.2	6.2
Total desmids			2.5		
Closterium moniliforme			2.5		
Cosmarium reniforme					
Total chrysophyceae					
Dinobryon					
Total blue greens	1.2		1.2		
Filamentous	1.2		1.2		
Invertebrata					
Rotifers					
Ciliates	1.2				
Nematodes	101.2	611.2	1026.2	81.2	63.7

Appendix 17. Major taxa of microalgae and invertebrates from Pond D (organism x 10^6 m^{-3}).

Taxa \ Time	0700	0800	1000	1100	1600
Total chlorophyceae	135.0	12.5	11.2	6.3	61.1
Filamentous	3.7		1.2	2.5	1.2
Flagellate	131.2	10.0		1.2	58.7
Chloroccus					
Pandorina			5.0		
Scenedesmus		2.5	5.0	2.5	1.2
Total diatoms	22.5	8.7	63.9	33.7	22.5
Total centrales			6.2		2.5
Total pennales	22.5	8.7	58.7	33.7	20.0
Cyclotella					
Melosira distans			6.2		2.5
Diatoma			33.7	6.2	2.5
Eunotia	3.7	2.5	5.0	1.2	
Frustulia vulgaris					
Gomphonema		1.2		12.5	6.2
Meridion circulare					
Navicula	10.0	2.5	10.0	3.7	
Pinnularia					
Synedra			2.5	3.7	1.2
Tabellaria flocculosa (long type)	1.2				
T. flocculosa		1.2			
Total desmids			2.5	6.2	
Closterium moniliforme					
Cosmarium reniforme					
Total chrysophyceae					
Dinobryon					
Total blue greens					
Filamentous					
Invertebrata					
Rotifers					
Ciliates					
Nematodes					