Journal of Plankton Research Vol.19 no.5 pp.647–654, 1997

SHORT COMMUNICATION

Bacteria–chlorophyll relationships in Ethiopian lakes of varying salinity: are soda lakes different?

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Abstract. Some soda lakes in East Africa are extreme with respect to their high abundance of bacteria and phytoplankton. We used regression analysis of 52 samples from 18 lakes in Ethiopia to demonstrate that soda lakes conform to a different relationship between bacterial abundance and phytoplankton biomass. The exponent of the power function relating bacteria and chlorophyll is much steeper for saline lakes, although still less than one. This may reflect that bacteria in eutrophic soda lakes are more substrate limited, and less controlled by grazers..

Several authors have attempted to generalize about the significance of bacteria in planktonic systems by looking at their quantitative relationship with chlorophyll or primary production (e.g. Aizaki *et al.*, 1981; Linley *et al.*, 1983; Bird and Kalff, 1984; Cole *et al.*, 1988). The analysis of bacteria and chlorophyll carried out by Bird and Kalff (1984) found saline Lake Elmenteita in Kenya to be extreme in both its chlorophyll concentration and its abundance of bacteria. It is also substantially above the common regression line; an apparent outlier which they omitted from the analysis based on its strong influence. Many of the highest values for phytoplankton biomass and productivity occur in East African lakes with high salinity as sodium bicarbonate (e.g. Talling *et al.*, 1973), and these soda lakes have also been noted for their extremely high bacterial abundance (Kilham, 1981). Lake Awassa, Ethiopia, is also in the Rift Valley, but of moderate conductivity and algal biomass. We reported earlier (Zinabu and Taylor, 1989a,b) that this lake does not have a particularly high bacterial biomass or production relative to its phytoplankton population.

The goal of this contribution is to assess the possibility that the relationship between bacterial abundance and chlorophyll is different in saline and freshwater lakes by examining the relationship among Ethiopian lakes of different salinity. [Saline lakes in Ethiopia have sodium as the predominant cation and bicarbonate/carbonate as the major anion; see Kebede *et al.* (1994).] Ethiopian lakes are particularly appropriate to examine in this regard, as they exhibit a wide range of salinity and chlorophyll concentration, and these two parameters are not strongly correlated.

A total of 18 lakes (Tables I and II) were visited 1–5 times between 1989 and 1993, and water samples were collected from the surface away from shore with acid-washed polyethylene bottles. Samples from the same lake were taken in different seasons or years to ensure that they were not temporally autocorrelated. The total number of samples used in our analyses was 52. The physical features of these lakes are provided in Table I. Although the number of lakes we were able

Table I. Locations and morphometric features of the lakes. Lakes Ardibo, Budameda, Koriftu, Mechaferra and Tilo have been omitted due to lack of data

Name	Area (km^2)	Altitude (m)	Z_{mean}	Z_{max}
Abijata	176	1578	7.6	14
Arenguadi	0.54	1900	5.5	19
Awassa	90	1680	10.7	23
Babogaya	0.58	1870	38	65
Bishoftu	0.93	1870	55	87
Chamo	551	1233	nd	13
Chittu	0.8	1600	nd	21
Hayq	23	2030	37.4	88
Hora	1.03	1850	17.5	38
Kilole	0.77	2000	2.6	6
Methara	3.2	1200	nd	nd
Shalla	329	1558	87	266
Zwai	442	1636	2.5	8

nd, no data.

to study is much less than those reported in studies that survey the literature, our study has the advantage that all of the data are derived from the same methods and all of the bacterial counts are by the same operator (G.M.Z.). These sources of error can be substantial (Bird and Kalff, 1984).

Lakes Abijata, Awassa, Chamo, Shalla and Zwai are large Rift Valley lakes. All have been the subject of earlier limnological investigations (see Kebede *et al.*, 1994). Lake Hayq is in the northern highlands, on the western edge of the rift valley. Smaller Lake Ardibo drains into Lake Hayq from the south via a small stream. Descriptions of these lakes and citations to earlier studies can be found in Kebede *et al.* (1992). Lakes Babogaya, Bishoftu, Arenguadi, Kilole, Koriftu and Hora are crater lakes south of Addis Ababa near Debre Zeit. Lake Chittu is a crater lake adjacent to Lake Shalla. Lake Methara is a shallow, lowland basin east of the other lakes. Limnological information concerning these, and citations to earlier literature, can be found in Green and Mengistou (1991). Budameda, Tilo and Mechaferra are crater lakes west of Lake Awassa. No previous limnological studies have been published on these.

Samples for bacterial counting were fixed in borax-buffered formalin (2.5% final concentration) at the sampling sites. These samples were kept at 4°C as much as possible and counting was carried out within 3–5 days. Water samples for chlorophyll analysis were stored in a dark cooler at ambient temperatures until they could be filtered and refrigerated within 1–8 h of collection.

Bacteria were counted by the acridine orange direct count method of Hobbie *et al.* (1977). Fixed bacterial samples were vortexed and 0.1–1.0 ml of the sample was mixed with 7–9 ml of pre-filtered water to ensure even distribution of cells on a pre-stained 0.2 μm pore size Nuclepore polycarbonate filter that had been pre-stained with irgalan black. Enumeration of bacteria was carried out with a Nikon Labophot epifluorescence microscope. At least 200 bacteria were counted from each of two duplicate slides.

Chlorophyll *a* was estimated spectrophotometrically on samples collected on Whatman GF/C glass fibre filters. Filters were extracted overnight with 90% cold

Table II. Abundance of bacteria, chlorophyll *a* concentration and conductivity for the study lakes. Data include the month and year when the samples were taken. Estimates are in parentheses

	Date (month/year)	Bacteria (millions ml^{-1})	Chlorophyll *a* ($\mu g\ l^{-1}$)	Conductivity ($\mu S\ cm^{-1}$)
Abijata	mean	43.8	101	28 685
	3/91	52.9	134	28 050
	5/93	26.4	49	31 280
	10/93	52.0	121	26 726
Ardibo	3/89	9.4	8.0	593
Arenguadi	mean	109.2	422	6039
	2/90	100	366	5554
	7/91	117	285	6324
	4/92	111	616	6240
Awassa	mean	6.4	22	837
	10/90	8.7	23	874
	3/91	4.9	23	827
	5/91	4.9	18	867
	7/91	7.0	26	874
	10/91	7.9	22	816
	5/93	5.4	21	820
	10/93	5.8	24	780
Babogaya	mean	9.7	33	844
	2/90	9.1	37	819
	7/91	8.2	29	832
	4/92	11.9	33	882
Bishoftu	mean	18.5	51	1605
	2/90	24.0	74	1591
	11/90	12.5	19	1591
	4/92	18.9	60	1632
Budameda	4/89	9.2	32	7344
Chamo	mean	16.7	111	1296
	6/89	24.5	262	1350
	8/90	19.5	181	(1350)
	3/91	17.4	45	1320
	11/91	13.8	59	1428
	8/92	19.8	123	1100
	1/93	18.1	97	1200
	2/93	12.7	86	1326
	10/93	8.0	40	1350
Chittu	3/91	48.4	224	48 000
Hayq	3/89	12.2	13	912
Hora	mean	21.0	36	2309
	2/90	29.0	29	2201
	11/90	18.5	29	2309
	7/91	17.6	56	2417
	4/92	18.8	30	2309
Kilole	mean	5.2	33	257
	7/91	5.7	35	278
	4/92	4.7	30	235
Koriftu	mean	22	101	270
	7/91	24.6	65	270
	4/92	19.1	136	270
Mechaferra	3/89	116.5	296	33 120
Methara	3/91	6.3	29	7441
Shalla	mean	4.4	7.1	23 450
	7/89	4.8	1.4	(23 450)
	3/91	5.1	17	22 080
	10/93	3.2	3	24 820
Tilo	4/89	41.1	53	15 120
Zwai	mean	11.3	94	362
	7/89	11.3	120	(362)
	3/91	16.0	154	410
	7/91	16.3	89	(362)
	10/91	9.8	99	374
	3/93	7.9	70	396
	5/93	9.1	81	295
	10/93	8.7	48	333

methanol in the dark. The abbreviated formula of Talling and Driver (1963) was used to calculate the concentration of chlorophyll *a*. No correction was made for phaeophytin.

Conductivity was measured on the lake with a YSI Model 33 meter. For some analyses, lakes were sorted as freshwater ($K_{25} < 1000$ µS cm^{-1}), moderately saline ($1000 < K_{25} < 10\,000$) or saline ($K_{25} > 10\,000$). These boundaries are slightly higher than the boundaries at 600 and 6000 used by Talling and Talling (1965), but appeared to us to be more consistent with the natural discontinuities in our data. We will return to this question after examining our data below.

The abundance of planktonic bacteria varied from 3.15 million to 117 million cells ml^{-1}, and chlorophyll concentration varied from 1 to 600 µg l^{-1} (Table II). The highest bacterial abundances were found in lakes Mechaferra and Arenguadi. The lowest abundance was found in Lake Shalla. All of these are soda lakes.

Curvilinear regression demonstrated a strong relationship between bacterial abundance and chlorophyll concentration (Figure 1, Table III). The slope is less steep (0.593 ± 0.143) than that (0.844 ± 0.093) reported by Bird and Kalff (1984). However, the latter slope is less when Lake Elmenteita was excluded (0.776 ± 0.094). When the same analysis is applied to saline lakes, moderately saline lakes and freshwater lakes separately, the strength of the relationship, as indicated by the coefficients of determination and slopes (exponents), is largest for saline lakes and smallest for freshwater lakes (Figure 2, Table III). The standard errors of the estimates indicate that the lower coefficient of determination for freshwater lakes

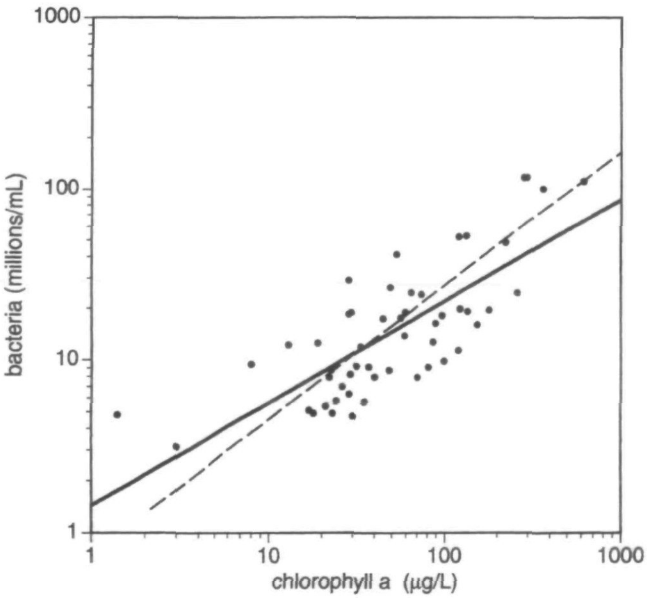

Fig. 1. The relationship between the abundance of bacteria and chlorophyll *a* concentration in 52 samples from 18 Ethiopian lakes. The solid line indicates the regression line from Table III and the broken line indicates the regression line from Bird and Kalff (1984).

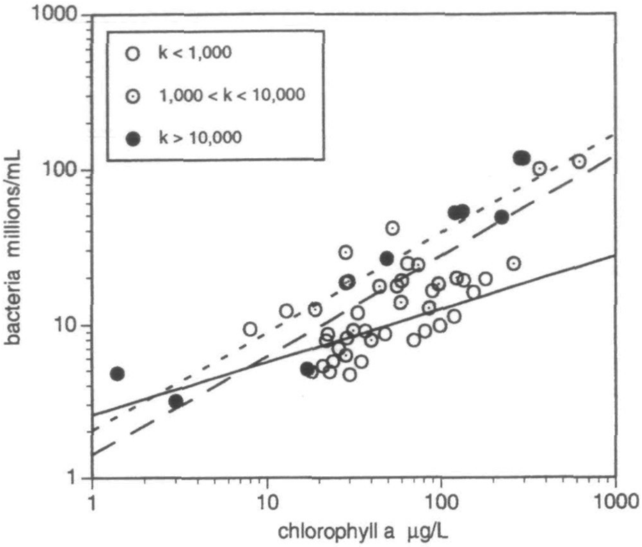

Fig. 2. The same data as for Figure 1, with separate regressions for freshwater ($K < 1000$), moderately saline ($1000 < K < 10\,000$) and saline lakes ($K > 10\,000$).

is because the variation in bacterial concentration is less, not that the scatter about the regression line is greater. The slope for freshwater lakes is significantly smaller than that for the other two lake groups (ANCOVA, $P < 0.05$). Freshwater and some moderately saline lakes with high chlorophyll concentrations had lower bacterial densities than expected.

We assessed the impact of using several data points from some lakes by repeating our analysis with lake means only. The result is very similar (Table III).

The importance of salinity can be assessed in a continuous fashion by using it as an additional independent variable in a multiple regression. The addition of this variable has little effect on the regression coefficient for chlorophyll, but it has a significant impact on the coefficient of determination (Table III). The samples from saline lakes with high chlorophyll concentrations are much above the regression line for freshwater lakes (Figure 3). However, five samples from lakes Shalla,

Table III. Regression statistics concerning the relationship between bacteria and chlorophyll in Ethiopian lakes. All variables are transformed as $\log_{10}$. Bacteria (bact) are expressed as numbers ml^{-1}, chlorophyll *a* (chlor) is expressed as $\mu g\ l^{-1}$ and conductivity (cond) is expressed as $\mu S\ cm^{-1}$

Dependent variable	Equation	n	r^2	$s(y)$
log(bact, all data)	0.156 + 0.593 × log(chlor)	52	0.57	0.252
log(bact, freshwater)	0.412 + 0.343 × log(chlor)	24	0.34	0.164
log(bact, moderate)	0.101 + 0.660 × log(chlor)	19	0.61	0.222
log(bact, saline)	0.323 + 0.643 × log(chlor)	9	0.85	0.219
log(bact, lake means)	0.040 + 0.700 × log(chlor)	18	0.65	0.245
log(bact, all data)	–0.627 + 0.590 × log(chlor) + 0.242 log(cond)	52	0.74	0.196

Methara and Budameda with high conductivity (all >7000), but low to moderate chlorophyll concentrations (≤30), conform to the freshwater pattern and appear to the lower right of Figure 3.

A hypothesis compatible with these data is that bacterial populations in the plankton of eutrophic freshwater lakes are suppressed by grazers, and do not increase in a manner proportional to algal biomass. This has been suggested by Bird and Kalff (1984) and others. That they increase at all may reflect changes in the threshold for grazer functional response and/or that particle-associated bacteria may continue to increase with surface area of larger particles, chiefly algae. In contrast, grazer control may be weaker in soda lakes, allowing bacteria to increase more steeply with algal biomass. In oligotrophic and mesotrophic saline lakes, the bacterial population is not markedly different from that in freshwater lakes, perhaps indicating that the level determined by grazers is not very different from the levels that would be set by substrate concentration.

This interpretation of our data is consistent with several published observations. First, freshwater (and marine) bacteria generally respond strongly to grazer removal (positively) and antibiotics (negatively), indicating a strong effect of grazers. However, in oligotrophic lakes, this effect is not always observed (e.g. Pace and Funke, 1991) and nutrient additions may have a marked effect on this numerical response [e.g. Chrzanowski *et al.* (1995) and references therein]. In Lake Awassa, Ethiopia, bacterial populations responded to grazer removal and antibiotics in a manner consistent with strong bacterivory. Seasonal variation in bacterial abundance was related to phytoplankton biomass, but with a lag that weakened the correlation (Zinabu and Taylor, 1989b). Similar experimental work has not been conducted on African soda lakes.

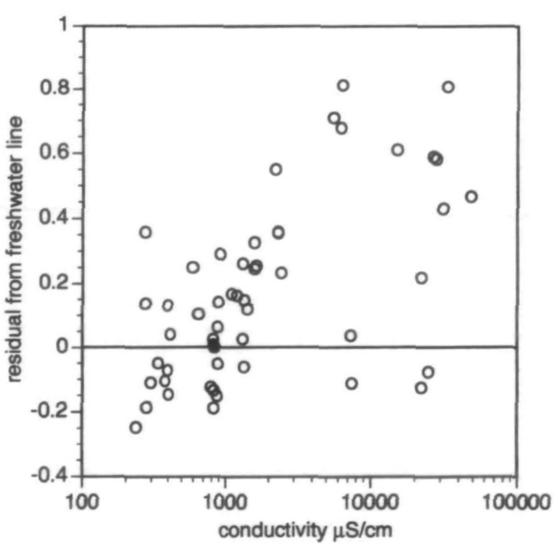

Fig. 3. Residuals from the regression for freshwater data only, calculated for all data and plotted against conductivity.

The relationship we observed among bacterial concentration, chlorophyll and conductivity is probably not unique to East African lakes. Campbell and Prepas (1986) found elevated bacterial abundance relative to the Bird and Kalff (1984) model in saline prairie lakes in Canada. In contrast, Robarts *et al.* (1995) measured bacterial abundance in a hypertrophic, but only weakly saline Canadian prairie lake, and found it conformed closely with the Bird and Kalff model. These limited data suggest that African and North American lakes of varying salinity may conform to a similar pattern.

If bacterial populations are not regulated by grazers in saline lakes, it remains to be determined why this is so. Ciliated protozoans may be very abundant in soda lakes (e.g. Finlay *et al.*, 1987; Yasinde, 1995), but these are not mostly bacterivores. We suggest that the flagellates that effectively graze bacteria in freshwater lakes and oceans (Fenchel, 1986) may not be abundant in these soda lakes. Our hypothesis that pelagic bacteria in saline lakes are substrate limited because protist grazers are absent can be tested with simple experiments that examine the numerical response of bacteria to filtration, dilution and antibiotics.

Acknowledgements

This research was supported by grants from the International Foundation for Science (to G.M.Z.), the National Science and Engineering Research Council of Canada (to W.D.T.) and the Canadian International Development Agency.

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Received on December 3, 1995; accepted on January 5, 1997