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Bacterial secondary production in oxic and anoxic freshwaters

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Abstract

Across a series of nine lakes that ranged from ultraoligotrophic to eutrophic, bacterial production was greatest in anoxic hypolimnetic water despite lower hypolimnetic temperatures. Mean (by lake) aerobic bacterial production ranged from 2.6 to 17 mg C $m^{-3}d^{-1}$ while mean anaerobic bacterial production ranged from 1.1 to 47 C mg C $m^{-3}d^{-1}$. Overall mean anaerobic bacterial production was 1.6 times greater than mean aerobic bacterial production. The relatively high rates of bacterial production in anoxic waters are due to larger bacterial populations growing slightly more slowly than their aerobic counterparts. Our data suggest that bacterial production in cold, anoxic water is as large or larger than that in warm, oxic surface waters and bacterial production in anoxic water is much greater than aerobic bacterial production at comparable temperatures.

We measured hypolimnetic accumulation of dissolved inorganic C to estimate the minimum possible growth efficiency for hypolimnetic bacteria. Growth efficiency, bacterial production/(bacterial production+bacterial respiration), ranged from 6 to 40% and averaged from 16 to 30% in anoxic water, depending on how it was calculated. The growth efficiency of anaerobic bacteria is about equal to that reported for aerobic bacteria.

A common observation is that sediments overlain by anoxic waters are rich in organic matter content. High deposition rates of organic matter from the water column may explain both the anoxia and the high organic content of the sediments (Henrichs and Reeburgh 1987), but anoxia is also believed to slow decomposition rates and facilitate the preservation of organic matter (Glenn and Arthur 1985), especially for certain classes of organic compounds (e.g. lipids; Harvey et al. 1986). Although a number of laboratory studies suggest little or no difference between oxic and anoxic decomposition rates (reviewed by Henrichs and Reeburgh 1987), evidence for retarded decomposition under anoxic conditions continues to be mixed. For example, Lee (1992) found little difference between the intrinsic rate of decay of small molecules in acrobic and anaerobic marine water at a variety of sites. On the other hand, comparing oxic and anoxic Black Sea water, Mopper and Kieber (1991) found much slower rates of microbial uptake for a suite of small molecules in the anoxic water. Although many compounds decompose more slowly under anoxic conditions some actually decompose more rapidly (Sun et al. 1993).

Microbial decomposition results in both the oxidation of some substrate and the conversion of some substrate into new microbial biomass. If substrates can be oxidized rapidly in anoxic environments, it is possible that the synthesis of new bacterial biomass (i.e. bacterial secondary production) is also rapid in anoxic environments. Only a few studies, however, have specifically compared bacterial production and doubling time in oxic and anoxic water columns (*see* Pedrós-Alió et al. 1993). Although typically cold and by definition devoid of oxygen, anoxic hypolimnia are also potentially rich locales for microbial growth. These environments are enriched in both organic substrates and inorganic nutrients, either of which may limit bacterial growth in surface waters (Pace and Funke 1991). Finally, both the diversity and abundance of bacteriovores are diminished in anoxic water which may lead to the accumulation of greater bacterial biomass (Fenchel et al. 1990; Cole et al. 1993).

We have previously documented that both bacterial abundance and cell sizes were consistently greater in anoxic relative to oxic waters in a series of 20 lakes (Cole et al. 1993). In this study we compare the production and doubling times of bacteria in anaerobic hypolimnia with that in oxic water across a gradient of nine lakes representing a range of trophic conditions.

Acknowledgments

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Table 1. Some characteristics of the lakes sampled. Chlorophyll *a* (μg liter^{-1}) and pH values are means of our measurements, April–August 1992. The presence or absence of anoxic bottom water is also shown.

Lake	Location	pH	Chl *a*	Anoxic bottom water
East Twin	42°01′N, 73°22′W	8.9	5.5	Yes
Giles	41°22′N, 73°05′W	5.5	0.7	No
Lacawac	41°22′N, 75°17′W	6.5	4.0	Yes
Minnewaska	41°43′N, 74°14′W	5.1	0.7	No
Mohonk	41°46′N, 74°09′W	7.8	4.9	Yes
Stissing	41°50′N, 73°37′W	8.3	8.2	Yes
Upton	46°33′N, 73°34′W	9.0	9.9	Yes
Waynewood	41°23′N, 75°21′W	7.9	4.5	Yes
Wononscopomuc	41°57′N, 73°26′W	8.8	3.9	Yes

Methods

We sampled three to five depth strata in nine lakes, each five times during April through August, generating a matrix of 207 samples of bacterial production and other variables. The lakes, all of which were thermally stratified, ranged from ultraoligotrophic to eutrophic and some had anoxic hypolimnia on several of the sampling dates. The lakes, shown in Table 1, have been more fully characterized in other studies (Caraco et al. 1991; Cole et al. 1993; Baines and Pace 1994). Dissolved oxygen was measured with a YSI digital meter; a reading of 0.1 mg liter^{-1} or lower was taken to be anoxic. Samples for dissolved inorganic C (DIC) were pumped directly into BOD bottles and transported to the lab on ice. DIC was measured by gas chromatography according to the method of Stainton (1973) using a Shimadzu GC-AIT gas chromatograph. Samples for chlorophyll *a* were passed through Whatman GF/F filters and frozen. Chlorophyll *a* was measured fluorometrically in methanol extracts (Holm-Hansen and Riemann 1978).

Bacterial production was estimated in all cases with the [^{3}H]leucine method (Kirchman et al. 1986), following the precautions described by Pedrós-Alió et al. (1993) for handling anoxic samples. In the field, water was pumped directly into 15-ml Corning centrifuge tubes and allowed to overflow three to five volumes before capping. At capping, the tubes were full and contained no visible bubbles. Samples were transported back to the lab in coolers near the environmental temperature. In the lab we added [^{3}H]leucine (sp act = 2.2 TBq mmol^{-1}) to a final concentration of 17 nM. Incubations were 0.75–1 h within $\pm$1°C of the environmental temperature. The extraction procedure and calculations followed that of Kirchman (1993). A sample consisted of five replicate incubations and a single time-zero control which served as the blank. To determine the leucine concentration that saturated leucine uptake, we added [^{3}H]leucine at a series of seven concentrations and incubated as described. In addition, we performed isotope dilution experiments for each lake and for both the epi- and hypolimnetic samples to estimate the contribution of unlabeled leucine to leucine uptake (Simon and Azam 1989). We express bacterial production as mg C m^{-3}d^{-1} and convert leucine uptake to C according to Kirchman (1993). Bacterial abundance was determined for each sample by the acridine-orange method (Hobbie et al. 1977) using an Olympus epifluorescent microscope at 1,200×.

Table 2. Isotope dilution (ID) factors and saturating concentrations for the leucine uptake measurements. Means ($\pm$ SE) for each grouping are shown (*see text*).

	N	ID factor	*N*	Saturating concn (nM)
Oxic	5	2.8±1.1	7	19.1±2.6
Anoxic	5	4.1±1.4	6	18.2±2.1

We report statistical comparisons between epilimnetic and hypolimnetic strata as well as between oxic and anoxic water. For overall comparisons we aggregated the data by strata and oxygen content and performed *t*-tests with $\alpha = 0.05$. We also performed two-way ANOVA to examine the effects of both system and oxygen status on bacterial production.

Results

Saturation and isotope dilution—Isotope dilution is the contribution of unlabeled leucine present in the water or synthesized during incubation which increases the pool of leucine beyond what was added by the investigator and is expressed as the ratio of total leucine to the added amount. We made 10 measurements of isotope dilution, five from oxic water and five from anoxic water, from seven of the study lakes. Isotope dilution factors ranged from 1.3 to 6.3 in individual trials (Table 2). The mean (with SE) isotope dilution factor for the entire data set was 3.5±1.4. The mean factor for oxic water was lower than the mean for anoxic water. These values are not statistically different from each other ($P > 0.05$), but both are higher than the value of 2 commonly used by other researchers (Simon and Azam 1989; Kirchman 1993). We have previously found dilution factors above 3 in the deeper, hypoxic waters of some lakes (Pace and Cole 1994).

We measured the concentration of leucine at which bacterial uptake was saturated 13 times, seven from oxic and six from anoxic water from seven of the lakes. Saturation varied somewhat (range, 12–34 nM) in individual trials but had nearly identical means in oxic and anoxic water (Table 2).

In our calculations we assume that saturation occurred at 17 nM and used an isotope dilution factor of 4 for all samples. The use of a single value for the dilution factor throughout the data set causes a potential overestimation of bacterial production in oxic waters relative to anoxic waters.

Bacterial abundance—Bacterial abundance averaged 1×10^{10} cells liter^{-1} and ranged ~100-fold from 5×10^{8} to 5×10^{10} cell liter^{-1} among all lakes and depths. There

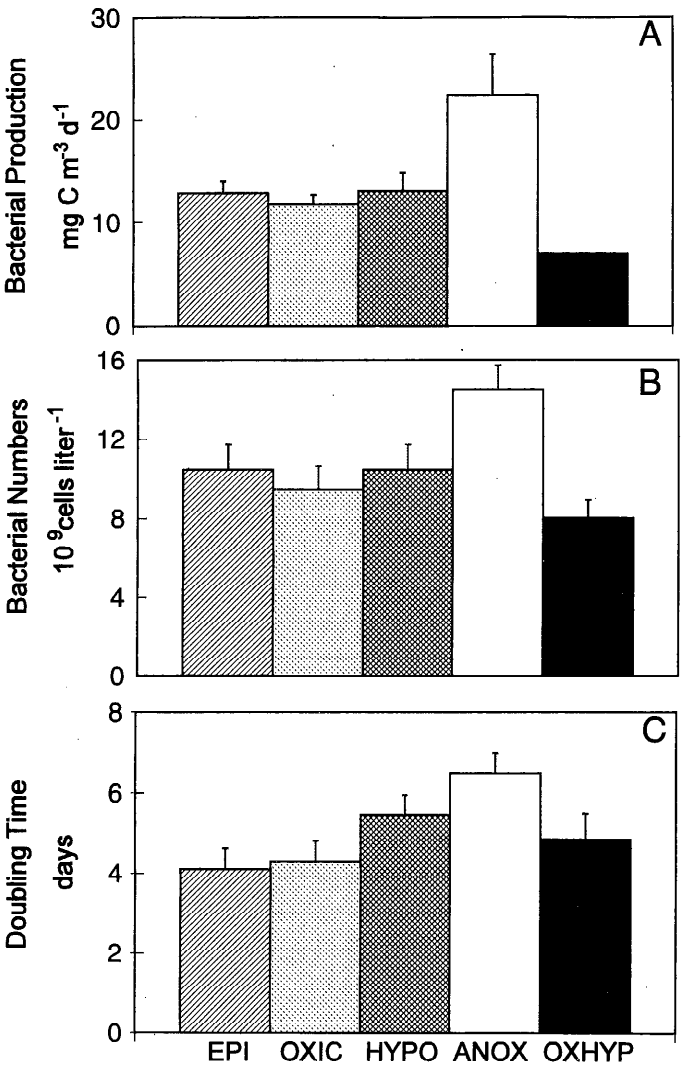

Fig. 1. Production (A), abundance (B), and doubling time (C) of bacteria from the water columns of the nine lakes studied. Means (with SE) are shown for all epilimnetic samples (EPI), all hypolimnetic samples (HYPO), all oxic samples from both the hypo- and epilimnia (OXIC), all anoxic samples (ANOX), and all oxic samples from the hypolimnia (OXHYP). For bacterial production and abundance, anoxic means were significantly ($P < 0.05$) higher than oxic means.

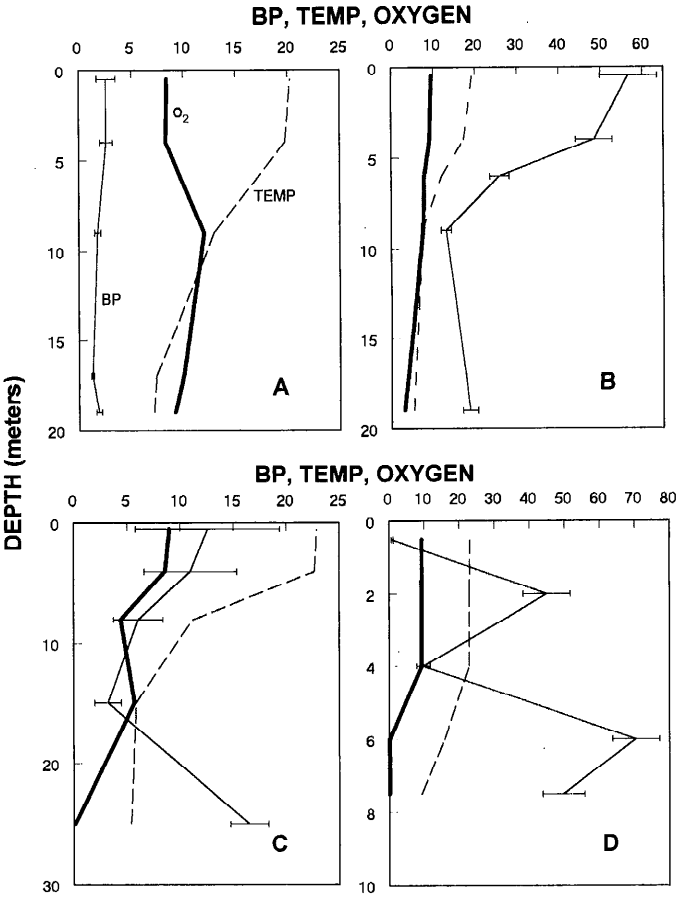

Fig. 2. Representative profiles of bacterial production (BP; mg C $m^{-3}d^{-1}$ [mean and SE]), temperature (°C), and oxygen (mg $liter^{-1}$) for four of the nine lakes studied. Two of the lakes (A—Minnewaska; B—East Twin) were oxic throughout at the sampling period; the other two (C—Wononscopomuc; D—Stissing) had anoxic hypolimnia.

were no overall differences between bacterial abundances in the hypolimnia and epilimnia across lakes ($P = 0.3$; t-test), but abundances were significantly greater in anoxic than in oxic waters ($P = 0.04$), consistent with other studies (Cole et al. 1993). A two-way ANOVA revealed a significant ($P < 0.01$) effect of both lake and anoxia on bacterial abundance. Further, all of the anoxic samples for bacterial abundance fell in the top quartile.

Bacterial production—Bacterial production among lakes, depths, and dates averaged 13.4 mg C $m^{-3}d^{-1}$ and ranged from 0.4 to 85 mg C $m^{-3}d^{-1}$. Once again there was no overall difference between mean bacterial production in hypolimnetic and epilimnetic samples ($P = 0.23$), but bacterial production from anoxic samples (mean 20.6 mg C $m^{-3}d^{-1}$) was significantly ($P < 0.05$) higher than that from oxic samples (12.6 mg C $m^{-3}d^{-1}$), despite the lower mean temperatures in hypolimnetic water (Fig. 1). A two-way ANOVA revealed a significant effect of both lake and anoxia on bacterial production ($P < 0.01$ in both cases). The interaction term (lake × anoxia) was also significant, indicating that bacterial production responds to anoxic conditions to different degrees in different lakes.

Bacterial production tended to be highest in either the surface waters or in anoxic bottom waters (Fig. 2). Comparing anoxic to oxic waters in each lake, the average ratio of bacterial production in anoxic to oxic waters in individual lakes ranged from 0.3 to 2.9 and averaged 1.6. We performed a series of t-tests comparing bacterial production in oxic to anoxic waters for each lake and for those dates for which we had anoxic waters (Table 3). Bacterial production was either significantly higher ($P <$

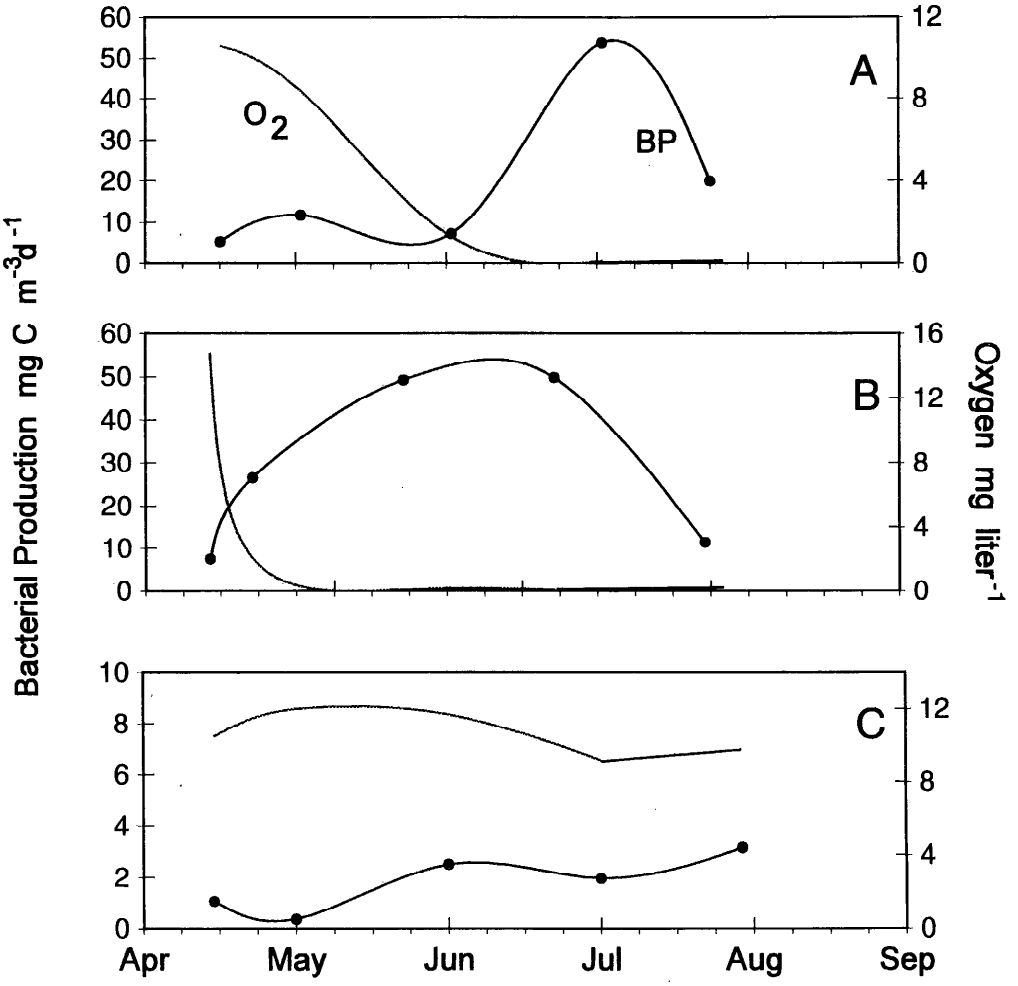

Fig. 3. Representative time-courses of dissolved oxygen (——) and bacterial production (—●—) in the hypolimnion of three lakes; A—Mohonk; B—Stissing; C—Minnewaska (Table 1). In Mohonk and Stissing anoxic conditions occurred; in Minnewaska, the hypolimnion remained oxic. Note different *Y*-scales.

Table 3. Mean anoxic and oxic bacterial production (ABP and OBP) in seven lakes. (Seven of the nine lakes we studied had anoxic water.) Shown are the difference between mean ABP and mean OBP (mg C m^{-3} d^{-1}) in each lake, the ratio of mean ABP to mean OBP, and the *P* value for a *t*-test comparing mean OBP and mean ABP in each lake. In no case is OBP significantly greater than ABP. Asterisks: *—$P < 0.05$.

Lake	ABP − OBP	ABP : OBP	*t*-test *P* value
East Twin	−9.3	0.38	0.07
Lacawac	2.3	1.90	0.04*
Wononscopomuc	7.3	1.90	0.19
Waynewood	10.3	3.20	0.02*
Upton	13.5	1.90	0.04*
Mohonk	20.0	3.90	0.11
Stissing	29.0	2.80	0.02*
Mean (SD)	13.7 (8.9)	2.30 (1.1)	

0.05) in anoxic than oxic (four of seven cases) or not significantly different from it (three cases; Table 3). In no case was bacterial production significantly greater in oxic water than it was in anoxic water. Further, 40% of the measurements of bacterial production in anoxic waters were in the highest quartile of all bacterial production measurements. Had we used the measured values of isotope dilution instead of a single value of 4 throughout, bacterial production in anoxic waters would average 3.3 times higher than bacterial production in oxic waters. In bottom waters that were oxic, bacterial production tended to be lower than it was in any other samples (Fig. 2). Over time, bacterial production tended to peak in anoxic waters soon after they had become anoxic and then to gradually decline (Fig. 3).

Growth rate—We express the specific growth rate of bacteria as the amount of time required to replace bacterial biomass. This rate is equivalent to doubling time and is equal to bacterial biomass divided by bacterial

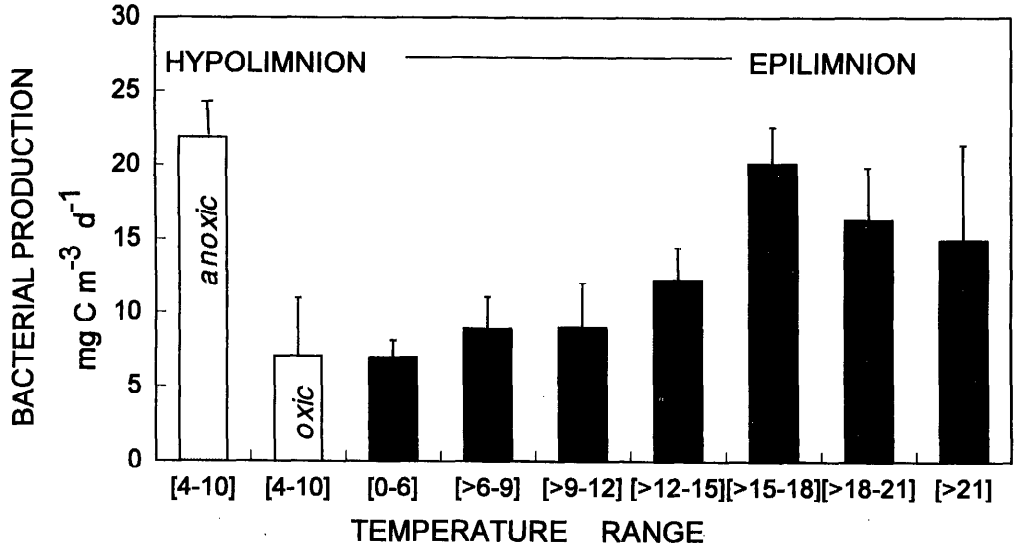

Fig. 4. Relationship between bacterial production and temperature aggregated by temperature range. We divided the data set into a series of temperature ranges and calculated mean bacterial production (with SE) for each range. The means for both oxic and anoxic hypolimnetic samples are indicated separately. Excluding the anoxic samples, the data fit a sigmoidal relationship, bacterial production = $35/\{1 + [\exp(2 - 0.1 \times \text{temp})]\}$, which accounts for 70% of the variance in oxic bacterial production. Anoxic bacterial production falls significantly above this relationship.

production. Doubling time among lakes and depths averaged 4.6 d and ranged from 0.15 to 34 d. Doubling time is significantly more rapid in water warmer than 15°C (3.6 d) than it is in water cooler than 15°C (5.1 d; $P = 0.05$), combining both oxic and anoxic data. At comparable temperatures, bacterial doubling time is significantly (*t*-test; $P < 0.05$) slower in anoxic water than it is in oxic water. The maximum temperature for anoxic samples was 10°C. Doubling time for oxic samples <10°C was 4.3 d; for anoxic samples, doubling time was 6.4 d.

At the scale of individual samples, temperature was not well related to bacterial production, doubling time, or to the log-transformed values of these variables ($r^2 < 0.1$; $P \gg 0.05$). Apparently other regulatory factors dominate and obscure a clear relationship with temperature. A relationship between temperature and bacterial production becomes apparent, however, by partially aggregating the data set. When mean bacterial production is blocked by temperature range, excepting the anoxic values, bacterial production increases with temperature up to ~15°C and then levels off (Fig. 4).

Discussion

Bacterial production was consistently high in anoxic water despite the low temperatures (<10°C) of hypolimnetic water in the lakes during the study period. Rates of bacterial production were highest either in the surface samples of the more eutrophic lakes or in anoxic hypolimnia. Similar trends have been observed for some but not all systems previously studied. For example, in Lake Ogelthorpe (McDonough et al. 1986), Big Soda Lake (Zehr et al. 1987), and Paul Lake (Pace and Cole 1994), the highest rates of bacterial production were observed just as oxygen concentrations became undetectable; bacterial production declined deeper into the more reduced hypolimnion. In Mirror Lake, Ochs et al. (1995) found bacterial production in anoxic water to be ~10 times higher than that in the epilimnion during summer stratification. In Little Crooked Lake, bacterial production in anoxic water was as great or greater than it was in the oxic epilimnion (Lovell and Konopka 1985). On the other hand, in both the Black Sea (Karl and Knauer 1991) and Crooked Lake (Lovell and Konopka 1985) heterotrophic bacterial production was highest in the euphotic zone with no peaks in the anoxic zone. The high rates of bacterial production in anoxic water that we studied were not caused by proximity to the sediments or by cool temperatures—both correlates of anoxia. At the same distances from the sediments (2 m), bacterial production was lowest in cold hypolimnia that did not become depleted of oxygen.

Some of the differences among results from different studies may be methodological. Some anaerobic bacteria do not transport exogenous thymidine (Gilmour et al. 1990; *but see* Robarts and Zohary 1993) which may lead to low estimates of bacterial production in some anoxic waters (Garcia-Cantizano et al. 1994). We estimated bacterial production by the uptake of [^{3}H]leucine and saw consistently greater rates of assimilation in anoxic water.

In agreement with other observations (Lovell and Konopka 1985; McDonough et al. 1986; and Zehr et al. 1987), we found that bacterial production often peaked midway through the anoxic hypolimnion rather than in the deepest, most reduced, depths. Similarly, in some of the lakes

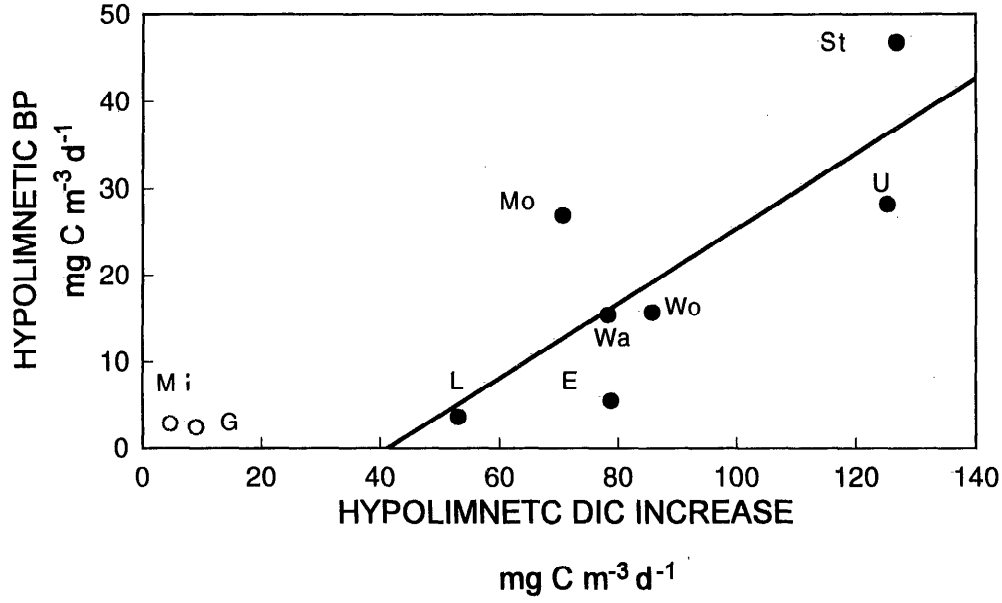

Fig. 5. An estimate of the growth efficiency of bacteria in the hypolimnion. The *X*-axis shows the measured hypolimnetic accumulation of DIC in each lake on a volumetric basis over the 112-d period of investigation. We use DIC accumulation as an estimate of bacterial respiration (*see* Caraco et al. 1989, 1991). The *Y*-axis shows mean hypolimnetic bacterial production (BP) estimated from leucine uptake. Anoxic hypolimnia—●; oxic hypolimnia—O. Key: Mi—Minnewaska; G—Giles; St—Stissing; U—Upton; Mo—Mohonk; Wa—Waynewood; L—Lacawac; Wo—Wononscopomuc; E—East Twin. The line is a regression of hypolimnetic DIC accumulation vs. hypolimnetic BP for the anoxic points only; it has a slope of 0.43 and an r^2 of 0.61 (*see text*).

bacterial production in the deepest depths peaked during early anoxia but declined over time throughout summer (Fig. 3). A similar pattern has also been reported for Little Crooked Lake (Lovell and Konopka 1985). Based on these patterns, we suggest that the lower rates of bacterial production observed in deeper anaerobic waters (or later in the season) are due either to the depletion of labile substrate or to the depletion of electron acceptors rather than to the effect of anoxia per se.

Several studies suggest that bacterial production increases with increasing temperature up to a threshold level at which other factors become limiting (White et al. 1991; Hoch and Kirchman 1993; Ochs et al. 1995). Because we studied several lakes with extremely wide-ranging conditions, a simple relationship between bacterial production and temperature is obscured, at least at the scale of the individual samples. By blocking the data into temperature ranges, we average much of this variation and observe a sigmoidal relationship between temperature and bacterial production (Fig. 4). This relationship is similar to that seen in both seasonal studies of some systems (above) and consistent with studies in which temperature is experimentally manipulated (Felip et al. 1995). It is intriguing that bacterial production in anoxic waters falls above this general relationship. Sun et al. (1993) found that the degradation of chlorophyll *a* was temperature-dependent in oxic waters but temperature-independent under anoxia. On the other hand, Cole et al. (1984) found that bacterial oxidation of algal organic matter in anoxic lake water was always enhanced by incubation at warmer temperatures.

Little is known about the growth efficiency of anaerobic bacteria in nature. An indirect, ecosystem-level approach is possible using the accumulation of DIC in the hypolimnion as a measure of hypolimnetic whole-system respiration, most of which is probably bacterial. That is, we followed the approach of Caraco et al. (1989, 1991) and used the accumulation of DIC in the hypolimnion as a measure of total bacterial respiration. We compared this estimate of bacterial respiration to our estimates of bacterial production based on leucine uptake. There is a reasonably good relationship ($r^2 = 0.61$) between measured bacterial production and the accumulation of hypolimnetic DIC among the systems we studied (Fig. 5). The slope of a plot of hypolimnetic bacterial production vs. hypolimnetic respiration is 0.28 across all systems (oxic and anoxic) and 0.43 in just the anoxic hypolimnia (Fig. 5). From these slopes, the computed mean bacterial growth efficiency, bacterial production/(bacterial production+bacterial respiration), would be 22% for all systems and 30% in anoxic water. If this same calculation is done system by system, the range is 6.6–39% and averages 17% for the anoxic systems. Since the accumulation of hypolimnetic DIC may be due to both water-column and benthic respiration and may be augmented by the redissolution of calcite precipitated from the epilimnion in

Table 4. Values of bacterial production (BP, mg C m^{-3} d^{-1}) in anoxic water columns. The name of the system along with the value and the citation are shown. Method: LEU—leucine uptake; ADN—adenine uptake; THY—thymidine uptake; FDC—frequency of dividing cells. Where the investigators did not provide data in the same units, we assumed that bacterial cells were 6×10^{-15} g C $cell^{-1}$ and the bacterial production continued at the hourly rate for 24 h d^{-1}.

System	BP	Method	Reference
	Anoxic waters		
Black Sea	<6	ADN	Karl and Knauer 1991
Lake Ogelthorpe	14–35	LEU	McDonough et al. 1986
Big Soda Lake			Zehr et al. 1987
Mixolimnion	5	THY	
Monimolimnion	0.4	THY	
Mirror Lake	31	THY	Ochs et al. 1995
Lake Tystrup	13	THY	Riemann 1983
Lake Ciso	170–340	FDC	Pedrós-Alió and Guerrero 1993
East Twin	5.6	LEU	This study
Lacawac	3.7	LEU	This study
Mohonk	27	LEU	This study
Stissing	46.8	LEU	This study
Upton	28.3	LEU	This study
Waynewood	15.5	LEU	This study
Wononscopomuc	15.8	LEU	This study
Mean (all anoxic)	30.6		
Mean (seasonally anoxic)	20.7		
	Oxic surface waters		
Mean from 54 studies	26.3	Mixed	Cole et al. 1988
Mean from 57 studies		Mixed	White et al. 1991
Freshwater	43		
Marine	17.9		
Estuarine	356		

some lakes, these estimates of growth yield must be considered minimal. In other words, if we assumed that the growth efficiency of anaerobic bacteria in the water column were only 10%, respiration of bacteria in the anoxic water column would more than account for the DIC buildup in most cases. Apparently the growth efficiency of these anoxic bacteria (16–30% depending on the calculation) is about as high as those estimates for aerobes in other studies (e.g. Cole et al. 1989; Kristiansen et al. 1992; Kroer 1993).

Combining our data with published estimates of bacterial production in anoxic waters suggests that, despite differences in techniques, the range of bacterial production from anoxic waters is about the same as that reported from oxic surface waters, excepting estuaries (Table 4). Clearly, bacterial production in anoxic waters is not lower than bacterial production in oxic waters and may indeed be higher, especially in seasonally rather than in permanently anoxic water.

At these rates, bacteria in anoxic water columns, not just those in the sediments, could be extremely important in the depletion of electron acceptors in the hypolimnion. If we assume that anoxic bacterial production is 19 μg C $liter^{-1}d^{-1}$ (the mean for seasonally anoxic waters), that bacterial growth yield is 30% (above), and that 1 mol of SO_4^{2-} can oxidize 2 mol of organic C, these bacteria have the potential to deplete the entire sulfate pool in ~20 d in low-sulfate soft-water (30 μM SO_4^{2-}) lakes or in ~80 d in typical hard-water lakes (120 μM SO_4^{2-}). Obviously SO_4^{2-} is not the sole available electron acceptor in anoxic freshwaters, but it is often much more abundant than nitrate, iron, or manganese and so we use it as an example. Thus, depending on the actual rate of bacterial respiration, sulfate concentration, and the presence of other electron acceptors, the continued growth of anaerobic bacteria could become limited by the lack of sulfate in a single season in freshwaters, resulting in a shift to methanogenesis as is commonly observed in sediments. In marine environments or saline lakes, depletion of water-column sulfate would not occur for decades at these rates.

Is there a sufficient input of organic C into the hypolimnion to sustain the rates of bacterial production and respiration we estimate? Baines and Pace (1994) measured the sinking flux of particulate matter out of the euphotic zone in a series of lakes, including seven of the nine lakes we studied. They developed a simple model relating the sinking flux of organic C to water-column chlorophyll concentrations. We used these equations and our measurements of chlorophyll to estimate the flux of carbon sinking into the hypolimnion of our study lakes and compared it to the estimated respiratory demand of the hypolimnetic water-column bacteria (Fig. 6). Using our measurements of bacterial production and assuming a growth efficiency of 30% (above), we see a reasonably

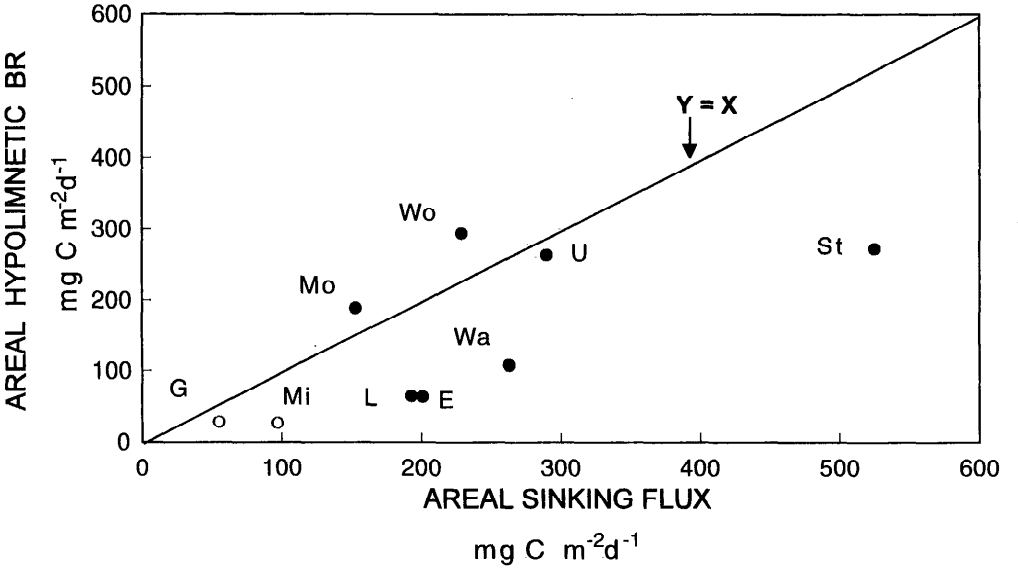

Fig. 6. Is the input of organic C by particle sinking sufficient to satisfy the C demand of hypolimnetic bacteria? The estimated sinking flux of organic C out of the epilimnion X-axis (*see text*) is plotted against our estimate of hypolimnetic bacterial respiration (BR) based on the leucine uptake measurements and an assumed growth yield of 30%. Both are expressed on an areal basis. The line $Y = X$ is shown for comparison. Symbols as in Fig. 5.

strong relationship between our estimates of bacterial respiration and sinking flux, both expressed on an areal basis ($r^2 = 0.46$; $P < 0.05$). The slope (0.55) suggests that about half of the particulate flux out of the euphotic zone is respired in these hypolimnia. Had we assumed a growth efficiency of 10%, bacterial respiration would be 2.1 times larger on average than the particle flux and could not be supported by particle flux alone.

Bacteria in anoxic water are doubling more slowly than those in oxic water at comparable temperatures. This somewhat slower doubling time is more than offset by greater bacterial numbers and biomass such that bacterial production in anoxic waters averages ~1.6 times greater than that in oxic samples despite the cooler temperatures. The slower doubling time of bacteria in anoxic water indicates that these bacteria persist in the environment longer than they do in oxic environments and may, as Lee (1992) suggested, contribute to organic matter preservation in anoxic environments. The role played by bacteria in the anoxic part of the water column is not well integrated into our thinking about either microbial ecology or lake ecosystem function. What limits the growth rates and abundances of these bacteria is not clear nor is the extent to which they are linked to other parts of the food web. Our study and others suggest that the magnitude of bacterial production in anoxic water columns is large enough to warrant further scrutiny.

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