Ecosystems (2004) 8: 1–14
DOI: 10.1007/s10021-003-0071-x

ECOSYSTEMS

Longitudinal Spatial Patterns of Bacterial Production and Respiration in a Large River–Estuary: Implications for Ecosystem Carbon Consumption

Roxane J. Maranger,[1†]* Michael L. Pace,[1] Paul A. del Giorgio,[2] Nina F. Caraco,[1] and Jonathan J. Cole[1]

[1]*Institute of Ecosystem Studies, P.O. Box AB, 65 Sharon Turnpike, Millbrook, New York 12545, USA;* [2]*Horn Point Laboratory, Cambridge, Maryland 21613, USA*

Abstract

Rivers and estuaries transport organic carbon (C) from terrestrial and freshwater ecosystems to the marine environment. During this transit, bacteria actively utilize and transform organic C, but few studies have measured detailed spatial variation in rates of bacterial respiration (BR) and production (BP). We measured BP at 39 stations and BR at 12 stations at monthly intervals along a 200-km reach of the tidal Hudson River. We observed strong repeatable spatial patterns for both BP and BR, with rates declining in the downstream direction. Bacterial Production had much greater dynamic range of spatial variation than BR. We used the detailed seasonal and spatial data on BP and BR to measure the total C demand of bacteria at several scales. We calculated volumetric and areal rates for 12 sections of the Hudson, as well as the total C utilization. Volumetric BR averaged 20 g C m^{-3} y^{-1}, but it was highest in the most upstream section at 30 g cm^{-3} y^{-1}. Areal rates averaged over the entire river were 174 g C m^{-2} y^{-1}, but they were 318 g C m^{-2} y^{-1} in the deepest section of the river, indicating the importance of morphometric variation. Total bacterial C demand increased downriver with increasing total volume. Overall, bacteria in the freshwater section of the river consumed approximately 18–25.5 $\times$ 10^9 g C y^{-1}, about 20% of the total organic C load.

Key words: spatial carbon cycling; bacterial respiration; bacterial production; river ecosystems; Hudson River.

Introduction

Rivers play a key role in the global carbon (C) cycle. A considerable fraction of terrestrial net ecosystem production is transported to the world's rivers (Schlesinger 1997). The quantity delivered to the ocean—some 0.4 Pt C y^{-1} (Aitkenhead and McDowell 2000)—is greater than the total amount of organic C buried in the world's oceans. Variability

*Corresponding author: Roxane J. Maranger; *e-mail:* RoxaneMaranger@aol.com or R.Maranger@umontreal.ca
Present address: Department of Biological Sciences, University of Montreal, CP 6128, Suc. centre-ville, Montreal QC, H3C 3J7, Canada
Present address: Department of Biological Sciences, University of Quebec at Montreal, CP 8888, Suc. centre-ville, Montrea QC, H3C 3P8, Canada
Received 7 May 2003; accepted 11 December 2003; published online ■.

in the sources of C in rivers impacts C budgets at both regional and global scales (Eckhart and Moore 1990; Clair and others 1994; Aitkenhead and McDowell 2000; del Giorgio and Duarte 2002). However, in addition to this transport function, rivers also degrade a large fraction of the organic C from both autochthonous and allochthonous sources. Much of this decomposition is due to the activity of bacteria.

Rivers are highly dynamic both spatially and temporally. Much of the prior work on bacterial production (BP) and bacterial respiration (BR) relies on temporally extensive but spatially limited sampling of river systems. So, although current estimates of riverine transport of organic C are probably reasonably accurate, the estimates of bacterial metabolism of organic C within rivers are far less certain because of this spatial undersampling.

Bacteria in rivers degrade organic matter originating from both aquatic and terrestrial sources. A large amount of allochthonous C enters from catchments providing supplemental organic C to within-system production, and large river systems tend to be net heterotrophic, wherein more C is respired than is locally produced (Cole and Caraco 2001). Net heterotrophy results in carbon dioxide (CO_2) evasion to the atmosphere and/or the export of waters with elevated dissolved inorganic carbon (DIC) concentrations relative to those upstream (Benner and others 1995; Raymond and others 1997; Raymond and Bauer 2001; Cole and Caraco 2001). Net heterotrophy may also be enhanced in turbid river systems where phytoplankton and macrophyte growth are limited by light penetration and deep mixing (Cole and others 1992).

Given that heterotrophic bacterial biomass and turnover rates are quite high in rivers, bacteria mediate large transformations of organic C in these and other aquatic ecosystems (Cotner and Biddanda 2002). Traditionally, studies of bacterial organic C transformations at the ecosystem level have focused primarily on bacterial C production (White and others 1991; Jahnke and Craven 1995). However bacterial C growth efficiency (BGE), which is the amount of C converted into biomass (bacterial production, or BP) relative to BP plus bacterial respiration (BR) (thus, BGE = BP:[BP + BR]), ranges from 5% to 60% averaging less than 30% across aquatic systems (Jahnke and Craven 1995; del Giorgio and Cole 1998). In terms of C cycling, BR is the more important term and must be included when considering bacteria in the context of ecosystem C fluxes (Jahnke and Craven 1995). Although more research has focused on BR in the last few years, there are fewer estimates of BR than BP because BR is difficult and time-consuming to measure.

The factors that influence bacterial C transformations in rivers are still poorly understood. Rivers are very dynamic and potentially subject to great spatial heterogeneity, making the analysis of regulatory factors more difficult. For example, point and nonpoint inputs are spatially heterogenous. Many rivers have significant,, temporally varying, exchanges with floodplains (Bayley 1995); and human activities influence the quality and quantity of nutrient and C inputs (Vörösmarty and others 1997; Caraco and Cole 1999, Rabalais 2002). Geomorphology also complicates analysis because rivers can vary considerably in width and depth over their length. Shoal areas may constitute zones of enhanced activity because of plant growth and benthic–pelagic coupling (Hopkinson and others 1998). All of these processes suggest that dynamic and variable microbial metabolism might be expected in rivers.

Past studies have most often considered only one or a few widely separated stations located longitudinally to estimate bacterial C metabolism in river ecosystems. However, the efficient methods currently available to measure BP and BR enable more samples to be processed in space in considerably less time. We conducted a detailed spatial sampling measuring BP at 5-km intervals and BR at 15-km intervals repeatedly along a 180-km transect of the Hudson River. The objectives of the study were to determine the variability of BP and BR along a river transect and to elucidate the factors that might be influencing any observed pattern. Another objective was to evaluate the importance of any variability in space and in time for determining an accurate estimate of bacterial ecosystem C demand.

Study Site

This study was carried out on the tidal freshwater and oligohaline portion of the Hudson River located in the state of New York, USA (Figure 1A). The Hudson River watershed is approximately 33,500 km^2. Land coverage is 67% forested, 23% agriculture, and 10% developed areas (Philips and Hanchar 1996). The tidal portion of the river, which we will refer to as the lower Hudson, is an eight-order tributary and is bounded to the north by a dam at Troy (referred to as river km [rkm] 250) and extends to the southern tip of Manhattan in New York City (rkm 0). Most of the fresh water (around 70%) entering the lower Hudson comes from the area drained north of the dam, with the balance from a number of tributaries further south

(Howarth and others 1996; Cole and Caraco 2001). Discharge averages around 350 m^3 s^{-1} at rkm 250 and average residence time is 0.15 y (Cole and Caraco 2001). However, in the year 2000, when our field study was undertaken, precipitation was elevated and average discharge was 440 m^3 s^{-1}.

From a morphometric perspective, the Hudson has extensive shoal (less than 3m) areas in the more northern reaches, and these account for approximately 35% of the area from rkm 160 to 250. From rkm 145 to 160, there is a gradual decline in the proportion of shoal relative to pelagic area from 35% to less than 10%; this proportion remains low for the rest of the tidal freshwater reach. Average river depth is around 9 m, but the river is considerably shallower in the north relative to the south (Figure 1B). The river is well mixed vertically (Cole and others 1992; Raymond and others 1997). The tidal amplitude ranges from 0.8 to 1.4 m across the entire tidally influenced freshwater section.

We sampled 39 stations, approximately 5–6 km apart in the central channel of the river, from rkm 232 just north of Albany to rkm 45 just north of New York City. The river is primarily tidal fresh water in this section, although salinity intrudes northward especially in late summer, when freshwater input declines. Samples were collected using an in-line flow system whereby water from an inlet pipe mounted exterior to the boat traveled through two large 30-L carboys. Water from the carboys was tapped into thoroughly cleaned polypropylene bottles while the boat was in motion. Over 20 stations were sampled in a 2-h period, and samples were kept in a cooler for approximately 2–4 h until analysis at the lab. We measured BP, bacterial abundance, chlorophyll-*a*, and nutrients from water samples collected at all 39 stations. Bacterial respiration and dissolved organic carbon (DOC) were measured at 12 of these stations, approximately every 15 km. Transects were carried out monthly from April until October 2000. We also conducted some preliminary transects measuring BP in late summer of 1999.

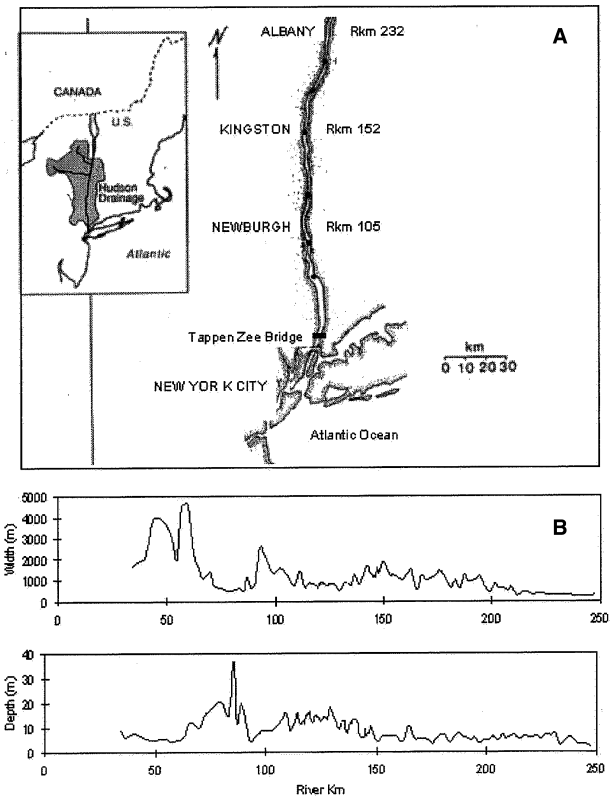

Figure 1. **A** Map of the lower Hudson River, New York. **B** Depth and width of the lower Hudson (m) along the transect studies. The x-axes in B represents station location in km, where river km (rkm) 250 is the dam at Troy to the north and rkm 0 is the southern tip of Manhattan Island.

Bacterial Production

Bacterial production was measured using the ^{3}H-leucine method (Kirchman and others 1985), as modified by Smith and Azam (1992). Water samples (1.5 ml) were dispensed, in triplicate, into clean 2-ml microcentrifuge tubes preloaded with 100 µl ^{3}H-leucine (44 Ci $mmol^{-1}$; New England Nuclear) to produce a final leucine concentration of 50 nM. Samples were incubated at in situ temperature for approximately 60 min. Rates of leucine incorporation were converted to bacterial C production (BP) using the conversion factors reported in Simon and Azam (1989). We confirmed that bacterial uptake of the final concentration of radioactive leucine was saturated (that is, maximum rates of uptake) by conducting a series of experiments in which increasing concentrations of ^{3}H-leucine were added to samples. We found that samples saturated at a leucine concentration of 50 nM. We verified previous results that additions of 50 nM of radioactive leucine to samples were diluted by a roughly equal concentration of unlabeled, background leucine (Roland and Cole 1999). We conducted isotope dilution experiments at five stations along the river transect to determine whether the in situ concentration of exogenous leucine varied in space. Briefly, BP was measured as above, in which the ^{3}H-leucine (50 nM final) added to samples was diluted with unlabeled leucine at six different concentrations (0, 42, 107, 159, 209, 259) (Moriarty 1986; Chròst 1990). The pool of in situ leucine was esti-

mated by plotting the inverse of the disintegrations per minute incorporated versus the concentration of unlabeled leucine added. The x-intercept of the least-squares regression of these two variables is equal to the amount of labeled ^{3}H-leucine added to the samples plus the in situ pool of unlabeled leucine diluting the ^{3}H-leucine.

Bacterial Respiration and Bacterial Growth Efficiency

Bacterial respiration was estimated by measuring the change in dissolved oxygen concentration in BOD bottles after dark incubation, as described in Roland and others (1999). Water samples were prefiltered (GF/D) to remove all grazers and enable almost all of the bacteria to pass into the filtrate. Differences in BP values between whole water and filtered samples suggested that on average less than 15% of the bacteria was removed. These were likely those bacterial cells attached to particles. Therefore, measured respiration rates may be underestimates. Twelve thoroughly cleaned 60-ml BOD were filled with filtrate; six were fixed immediately with Winkler reagents, and six were incubated in the dark from 20–32 h at in situ temperature. Most incubations were 24 h; however, when water temperatures were lower than 15°C, we extended the incubation time. After fixation of all bottles, 1 ml concentrated H_2SO_4 was added and the dissolved oxygen concentration was determined by a spectrophotometric modification of the Winkler technique (Duval and others 1974; Roland and others 1999). Periodically, dissolved oxygen concentrations that had been measured spectrophotometrically and calculated using the models provided in Roland and others (1999) were also verified via regular Winkler titration (Wetzel and Likens 1999). Oxygen concentrations calculated from the two methods were not significantly different.

Oxygen consumption (mg O_2 L^{-1} h^{-1}) was estimated from the difference in oxygen concentration between time zero and the end of the incubation over the incubation period. Values were converted to C respired (μg CL^{-1} h^{-1}), assuming a respiratory quotient of one. If the coefficient of variation among replicates was greater than 3% or if there was no measurable difference in the spectrophometric readings after incubation, the oxygen measurement was discarded. In total, three observations were discarded. To test that oxygen declines were linear, we ran a 24-h incubation sampling every 4 h for changes in oxygen concentrations. The decline in O_2 was indeed linear over the 24-h incubation, with an $r^2 = 0.96$. There was no apparent difference between the slope of the regression and the rate of oxygen consumption estimated from the two endpoints over time. The 95% confidence intervals of the slope of the regression and the rate of oxygen consumption estimated from endpoints overlapped.

Bacterial growth efficiency was calculated as the ratio BP:(BP + BR), thus estimating the proportion of C cycling through the bacteria that is actually converted into bacterial biomass. We used a mixed-model approach (Roland and Cole 1999) where the BP estimates were based on very short-term measures made on filtered water during the 1st hour of incubation and BR was measured during a 24-h incubation. This approach seems the most reasonable based on the assumption that short-term leucine uptake rates provide the best estimate of field rates and that respiration measurements were linear over the course of 24 h (see above).

Bacterial Biomass

Samples for bacterial abundance were fixed with glutaraldehyde (1%) and stored at 4°C until enumerated. Total bacterial abundance was determined by flow cytometry (del Giorgio and others 1996) using a FACScan (Becton Dickson, Mountain View, CA, USA) equipped with a 15-mW, 488-nm air-cooled argon-ion laser. Water samples (1 ml) were placed in cytometry tubes and stained with Syto-13 (Molecular Probes) at a final concentration of 1 μM, mixed with a Vortex mixer for a few seconds, and incubated for 10 min. Bacterial abundance determined via flow cytometry is based on the enumeration of bacterial particles relative to an internal standard. Samples were amended with pre-calibrated 1-μm fluorescent beads (Fluoresbrite Microspheres; Polyscience), and green fluorescence and side scatter were used to discriminate among stained particles, the bead standard, and noise.

Other Analytical Methods

Water samples for chlorophyll-*a* were filtered through a Whatman GF/F, and filters were frozen prior to analysis. Chlorophyll-*a* was extracted using methanol as described in Holm-Hansen and Reimann (1978) and read with a Turner Designs fluorometer. For dissolved nutrients, water samples were prefiltered through a 25-mm Gelman A/E filter in a thoroughly rinsed polypropylene

syringe and Swinex syringe filter tip. Nitrate plus nitrite were measured using the sulfanilamide method by reduction to nitrite with passage through a Cd column as modified for an Alpkem Autoanalyzer (APHA 1992). Greater detail for both chlorophyll and nutrient analysis are provided in Lampman and others (1999). Dissolved organic carbon was measured with a Shimadzu (Kyoto, Japan) 5050 carbon analyzer that uses high-temperature oxidation.

Bacterial Respiration and Carbon Demand

To estimate the volumetric and areal BR and bacterial C demand (BCD), the river was divided into 12 sections, approximately 15 km in length, centered by each bacterial respiration station. To calculate whole ecosystem estimates, we assumed that BP and BR did not vary significantly with depth or width and that the rates measured in the mid-channel were representative of the river section. Morphometric details of each section, including river volume, surface area and average depth, are well known. The monthly BCD per unit volume for each section was the sum of the calculated monthly mean in situ BP around each respiration station plus the average BR (BCD = BP + BR). Values from May until October were measured. The BCD was also estimated from four stations for the month of April 2000; these values were not significantly different from each other because no spatial pattern in BP or BR had emerged at that point in the season and a mean value for April was assigned to all stations. Annual BCD and BR for each river section was the sum of the monthly estimates. We assumed that from December until March, bacterial C consumption was negligible; estimates reported here are based on an annual growing season of 214 days. Total bacterial C requirements per section were estimated by dividing the volumetric BCD estimate at each section by the known volume.

For whole ecosystem estimates of volumetric BCD the following equation was used:

$$\text{Volumetric BCD (g C m}^{-3}\text{ y}^{-1}) = \sum_{n=1\ldots N}^{N} BCD_n \times \lambda_n \quad (1)$$

where n denotes the of river section, BCD_n is the estimated annual BCD per river section, and λ_n is the volumetric proportion of the section relative to the entire river reach. For areal estimates, the equation was modified to:

$$\text{Areal BCD (g C m}^{-2}\text{ y}^{-1}) = \sum_{n=1\ldots N}^{N} BCD_n \times z_n \times \gamma_n \quad (2)$$

where n denotes the river section, BCD_n is the estimated annual BCD, z_n the average depth per section, and γ_n is the areal proportion of the section relative to the entire river reach. Ecosystem level BR was estimated using the same equations and by substituting BCD with BR. Total C consumption (g C y^{-1}) was determined by multiplying volumetric BCD with the volume of the specific section.

Results

Spatial *in situ* Concentrations of Leucine and Isotope Dilution

The isotope dilution (ID) from all six sites ranged from 1.55 to 2.3, with an average of 1.9. There was no particular pattern north to south in the ID; therefore, we opted to use an ID of 2 for the entire data set, consistent with prior studies. This result indicates that in situ concentrations of leucine are about 50 nM.

Spatial Trends in Bacterial Metabolism

Initial transects carried out in the late summer of 1999 showed a distinct spatial pattern in BP where higher production values were observed in the northern segment of the reach above river km 150 (Figure 2A). Transects carried out the following year showed the same pattern (Figure 2B and C), even in different seasons. In general, BP was up to 10-fold greater in the most northerly stations near Albany, declined from rkm 200 to 150, and remained fairly constant below rkm 125. In contrast, patterns in BR were less consistent from month to month. Rates varied from fairly even along the entire transect in May 2000 (Figure 2B) or declined sharply from north to south in August 2000 (Figure 2C), mimicking the pattern in BP. Seasonally, rates of BR were variable at each station and were probably influenced by changes in temperature. Indeed, extreme high and low BR rates were observed in August at a river temperature of 23°C (mean, 5.66 μg C L^{-1} h^{-1}) and in October at a river temperature of 14°C (mean, 2.62 μg C L^{-1} h^{-1}). However, there was no correlation between average monthly BR and temperature, primarily because respiration was low during the July transect when temperature was very high (mean BR, 3.15 μg C L^{-1} h^{-1}; mean temperature, 24°C).

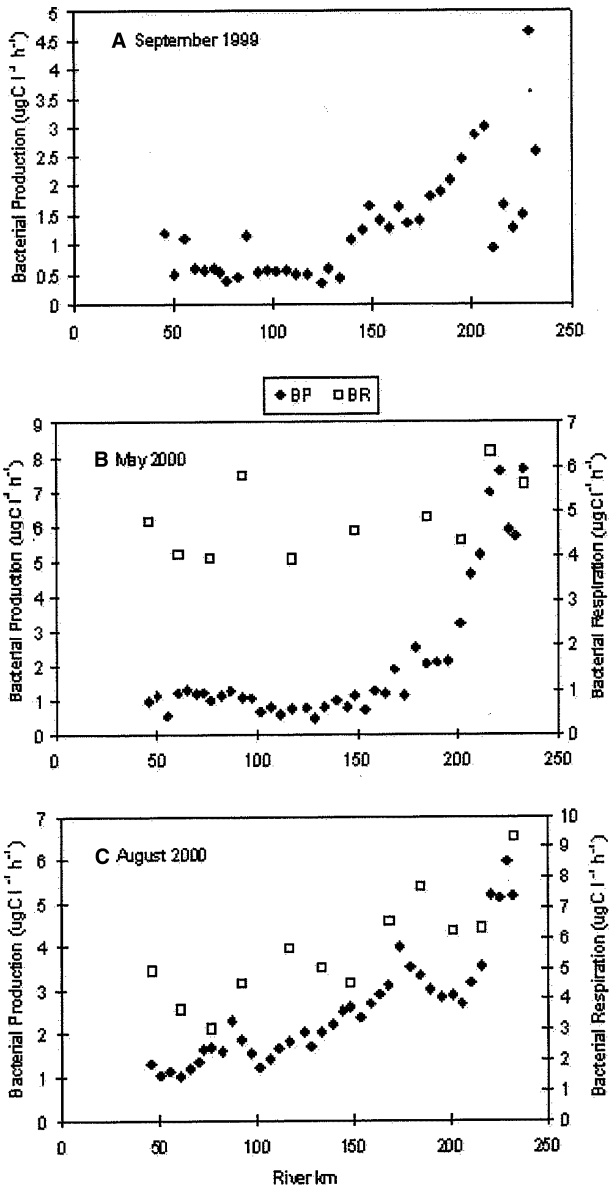

Figure 2. Pattern of bacterial production (μg C L^{-1} h^{-1}) along the transect of the Hudson River on various sampling dates. **A** September 1999. **B** May 2000. **C** August 2000. The x-axis represents station location in km, where river km 250 is the federal dam at Troy to the north and river km 0 is the southern tip of Manhattan Island. Pattern of bacterial respiration (μg C L^{-1} h^{-1}) along the Hudson River in May 2000 (B) and August 2000 (C).

The average rates of BP and BR from May through October 2000 along the transect are shown in Figure 3A and B. Spatially, BP rates were on average four-fold higher in the northern reaches of the river relative to the south, whereas rates of BR were less variable spatially, with rates less than one and a half fold greater in the north than in the south. Despite the spatial differences in the average rates of BP and BR, the coefficients of variation among stations were quite high (range and median coefficient of variation [CV] for BP, 27%–72%, 40%; range and median CV for BR, 8–52%, 35%), but CV did not vary in any consistent manner for both variables.

The higher rates of BP in the northern section of the river are reflected in an increase in BGE. A clear pattern in BGE is also apparent (Figure 3C), whereby the efficiency of C conversion into bacterial biomass was significantly higher in the north. Again, the CV for BGE was high (median CV, 30%; range, 23%–70%) and showed no pattern among stations. We also found a significant positive relationship between BP and total bacterial abundance ($r^2 = 0.49$, $P < 0.0001$, $n = 222$), Average bacterial abundance (BA) along the river transect is virtually identical to the consistent pattern observed for BP (Figure 3D).

Spatial variation in BP was not correlated with spatial variation in algal biomass. Chlorophyll-*a* was on average lowest at the most northerly stations and increased to a maximum around rkm 150 (Figure 4A). This pattern was quite distinct from the one observed for BP (Figure 3A). Bacterial production had distinct patterns in relation to inorganic nitrogen concentrations (Figure 4B). Indeed, there was a significant positive linear relationship between the mean rates of BP and the mean seasonal concentration of NH_4. Using ordinary least-squares regression analysis, the relationship is described by the equation $BP = 0.12 + 0.5\ NH_4$ ($r^2 = 0.55$, $n = 36$, $P < 0.0001$). The relationship between BP and the mean seasonal concentration of nitrate was negative and best explained using a curvilinear model: $\ln BP = 3.6–0.14\ \ln\ NO_3$ ($r^2 = 0.45$, $n = 36$, $P < 0.0001$). Bacterial Production was relatively invariant when NO_3 concentrations were greater than 35 μmol L^{-1}. For both the relationships between forms of inorganic nitrogen and BP, the most southerly three stations were eliminated from the analyses because of the influence on those stations of saline waters and nitrogen derived from sewage inputs from New York City. Average seasonal DOC concentration followed the same pattern as BP with an obvious decline downriver (Figure 4C).

Bacterial Carbon Demand in the Hudson River Ecosystem

In evaluating whole-ecosystem C requirements by the bacterial community, we present two values that represent alternative estimates of bacterial C

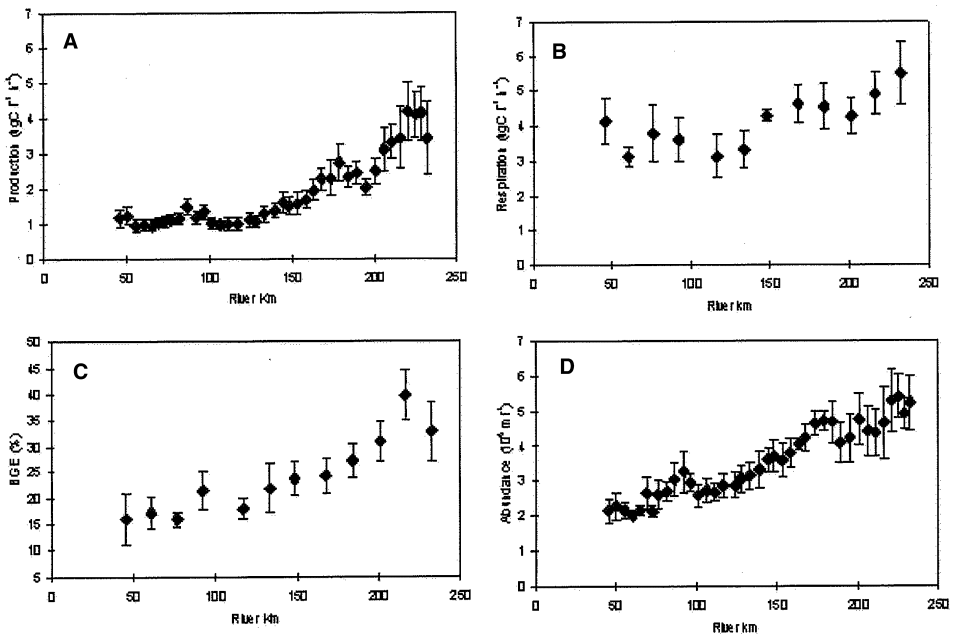

Figure 3. **A** Mean rate of bacterial production, **B** mean rate of bacterial respiration, **C** mean bacterial growth efficiency (BGE), and **D** total bacterial abundance from May to October 2000 along the Hudson River transect. Error bars represent the SD of the mean.

metabolism. Bacterial C demand (BCD) is the maximum C required by the bacterial community; it assumes that the fate of all BP is consumption and subsequent respiration by predators (not by bacteria). Bacterial respiration is a more conservative estimate of C demand and is based on the assumption that bacterial biomass production is ultimately remineralized by bacteria.

Although BR was quite stable in space, we wanted to determine whether BR and BCD were variable in time. Monthly volumetric ecosystem estimates for BCD and BR are shown in Figure 5. There was a twofold difference in the monthly bacterial C requirements between the extreme rates of the months of August and October, suggesting that some temporal variables are influencing the overall ecosystem C requirements. Although temperature would seem to be the most likely variable, we saw no clear trend with temperature. However, we did see an effect of discharge rate on ecosystem C consumption in which C requirements were highest in months with higher discharge rates. For the months of May, June, and August, the average discharge rates 1 week before the sampling date were greater than 360 $m^3 s^{-1}$ whereas the rates of discharge for July, September, and October were less than 225 $m^3 s^{-1}$. In a paired *t*-test of sample means, the rates of BCD and BR were higher in months with higher discharge rates than those with lower discharge rates (BCD, $t = 8.90$, $P < 0.01$; BR, $t = 8.92$, $P < 0.01$). These results suggest that sampling frequently during the course of a growing season is necessary to the accurate determination of annual ecosystem estimates.

Annual C requirements were determined in 15–20-rkm sections around each of the respiration stations along the river transect (Figure 6A–C), Volumetrically, BCD and BR were highest in the northern reaches of the lower Hudson (Figure 6A) at sites of highest BGE (see Figure 3C). Volumetric BCD and BR declined from rkm 158 until rkm 84 and began to rise again in the southern reaches at the sites where the lowest BGE were estimated (Figure 3B). Maximum values were approximately double the minimum for both BCD (range; 21–48 g C m^{-3} y^{-1}) and BR (range, 16–30 g C m^{-3} y^{-1}).

Areal estimates of BCD and BR were also determined for each of the 12 sections of the river (Figure 6B). Again, there was considerable variability among stations for both BCD (CV = 25%; range, 168–420 g C m^{-2} y^{-1}) and BR (CV = 31%, range, 127–317 g C m^{-2} y^{-1}). By considering the areal proportion that each river section represents, we estimated an annual ecosystem bacterial C respiration rate from rkm 247–38 of 174 gC m^{-2} y^{-1} for the year 2000.

In the year 2000, the total amount of C required for the bacterial community in terms of BCD and BR for the entire reach of lower Hudson was 53 and 39×10^9 g C y^{-1}, respectively. When broken down into the 12 river segments, total C requirements did not follow the areal or volumetric pattern in C demand but was a function of increasing river volume downstream (Figure 6C).

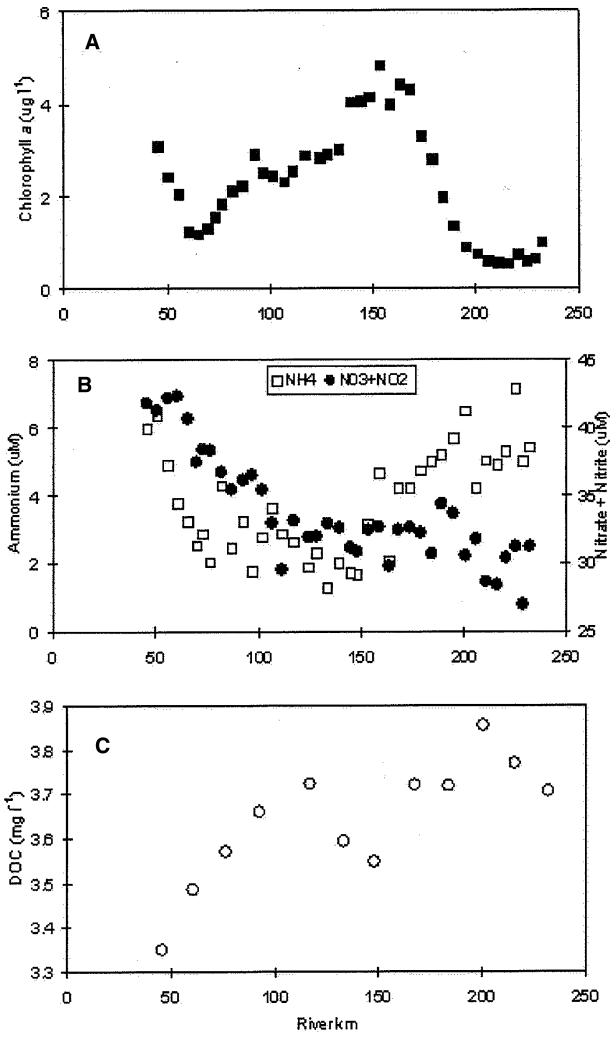

Figure 4. Mean seasonal concentrations of **A** chlorophyll-*a* (μg mol L^{-1}), **B** ammonium (μg mol L^{-1}), and nitrate (μg mol L^{-1}), and **C** dissolved organic carbon (DOC) (mg L^{-1}) from May to October 2000 along the Hudson River transect.

Discussion

The detailed longitudinal sampling of the Hudson River revealed a consistent spatial pattern in BP along a 180-km reach with higher rates in the north that declined as the river flowed south. From May to October 2000, the average rate of BP estimated from the station means for the entire river was 1.58 μg C $L^{-1}h^{-1}$, with a CV of over 80%, highlighting the important spatial variation in BP among sampling stations. In the case of BR, the trend was more constrained along the river transect. The average BR taken from the station mean values was 4.11 μg C L^{-1} h^{-1}, with a CV of 18%. Bacterial growth efficiency, like BP, consistently declined from north to south, with an average

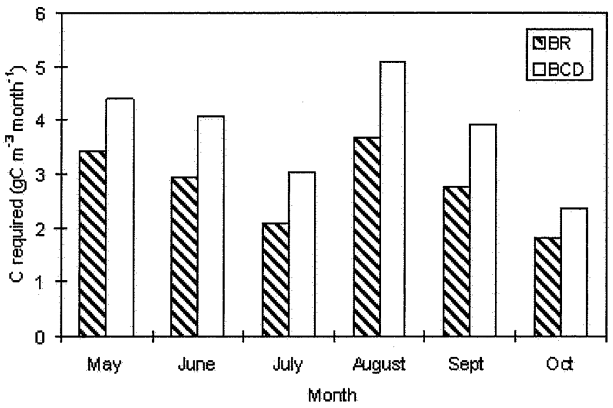

Figure 5. The volumetric bacterial carbon demand (BCD) and bacterial carbon respiration (BR) estimated on a per-month basis in the year 2000 for the entire river reach (rkm 45–247).

growth efficiency of 24% and a CV of 30%. Relative to BP, the CV of the BGE was much lower, suggesting that BP and BR were responding similarly to the different spatial and temporal conditions of the river. Indeed, BP and BR were related, but the relationship was weak (r^2 = 0.24) with a slope significantly lower than 1, supporting the previous conclusions that BP is the more dynamic variable (del Giorgio and others 1997; Cimbleris and Kalff 1998; Roland and Cole 1999; del Giorgio and Cole 2000). This study shows the relative importance of how these different bacterial metabolic processes vary along the flowpath of a large river ecosystem and evaluates the consequence of that variation for estimating ecosystem-level bacterial C demand.

Factors Influencing Spatial Variation in Bacterial Production and Bacterial Growth Efficiency

Higher rates of BP and BGE in the northerly reaches of the Hudson indicate that more labile organic C and/or nutrients are available to bacteria. What are the possible sources of microbial substrates that might fuel this enhanced activity and incorporation efficiency? The largest source of organic C (OC) loading into the river comes from nonpoint allochthonous watershed inputs (Table 1), with 70% of the total allochthonous load coming over the dam at Troy near the northernmost station we sampled. Phytoplankton and macrophyte primary production is less than (5% and sewage inputs were approximately 12% of the total allochthonous C load (Table 1). Allochthonous inputs are therefore far more important

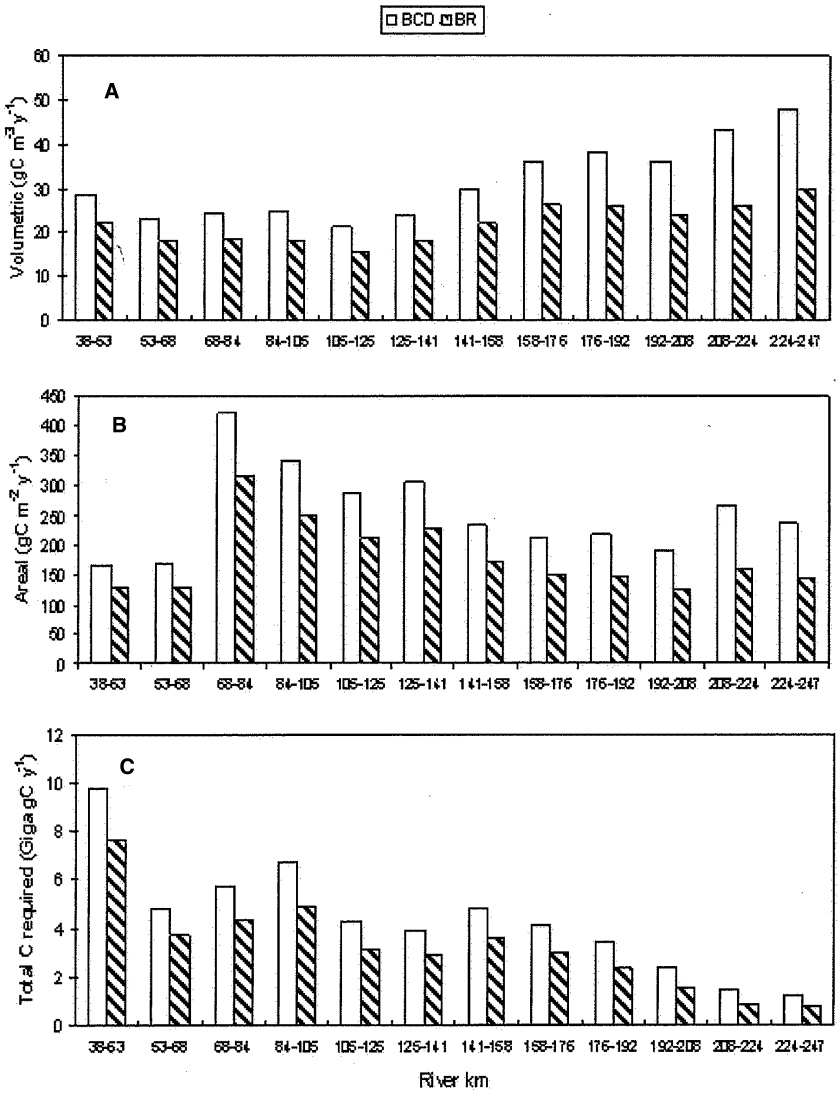

Figure 6. The bacterial carbon demand (BCD) and bacterial carbon respiration (BR) for denoted sections of the river in km **A** Volumetric (g C m^{-3} y^{-1}). **B** Areal (g C m^{-2} y^{-1}). **C** Total carbon (C) required (Giga g C y^{-1}). 16

quantitatively, but autochthonous C and sewage C likely provide more readily available substrates for bacteria.

Organic C from primary producers, including macrophyte leachate, is known to be labile and to favor higher rates of BP (Cole and others 1988; Wetzel 1992; Moran and Zepp 1997). In the case of phytoplankton, chlorophyll-*a* does not follow the pattern of BP, and a lack of coupling between phytoplankton and bacterial production has been previously described for the Hudson (Findlay and others 1991). Although there is greater macrophyte cover north of rkm 150 (16% relative to 7% below rkm 150), it should be noted that the sites with the most intense macrophyte production do not coincide with the main peaks in BP that we observed.

Another possible contributor to the increased rate of BP in the north is sewage input. Much of the load enters the river from the metropolitan Albany area (Figure 1). Sewage inputs can stimulate BP and create repeatable spatial patterns, as has been observed in the Seine (Servais and Gamier 1993). Sewage inputs might enhance BP and BGE both directly through labile organic matter and indirectly by providing inorganic nutrients, as evidenced by the more elevated concentrations of NH_4, that promote the degradation of the large amounts of allochthonous organic C entering the system from above the dam at rkm 250. Thus, the tidal Hudson might act like a flowthrough reactor where upstream, more labile inputs of organic C promote bacterial growth whereas, as lability declines downstream, BP stabilizes at a lower, consistent rate.

Spatial Stability of Bacterial Respiration

Average BR was quite conservative spatially relative to other variables. The observed lower variability in

Table 1. Estimates of Various Carbon Rates for the Hudson River

Variable	Areal C ($g\ C\ m^2\ y^{-1}$)	Total C ($g\ C\ m\ y^{-1}$)
Bacterial respiration	171	18
Bacterial production	70	7.5
Net primary production	31.4[c]	3.5
Net macrophyte PP	7.4	0.75
Allochthonous OC loading[a]	825	92.7
Autochthonous OC loading	38.4	4.25
Sewage inputs[b]	102	11.4

C, carbon; PP, primary production; OC, organic carbon
Unless specified, rates are for the year 2000. For consistency with other studies and to avoid uncertainties associated with inputs of marine C and sewage C from New York City, estimates are determined for the freshwater section from rkm 247 to rkm 105 (Albany to Newburgh, NY).
[a]*Estimated as the sum of average dissolved OC times the discharge rate and the average POC times the discharge rate measured at rkm 216 multiplied by 0.7 (70% of total allochthonous load)*
[b]*From Howarth and others (1996)*
[c]*Caraco and Cole (unpublished)*
[d]*Caraco and Cole (2002)*

BR as compared with BP in the Hudson has also been observed in both small-scale experimental manipulations (Roland and Cole 1999) and large-scale comparative studies (del Giorgio and others 1997). Indeed, the range in BR observed among systems is half of BP (del Giorgio and Cole 2000). Why is BR in aquatic systems apparently so constrained?

There is growing evidence that bacterial communities consist of many species in very different metabolic states (del Giorgio and Scarborough 1995; Williams and others 1998; Boulos and others 1999; Giovannoni and Rappé 2000; Maranger and others 2002). One plausible explanation as to why BP is more variable than BR is that all bacterial cells respire, but only a smaller and more variable fraction is actually dividing and accumulating biomass (del Giorgio and Cole 2000). Thus, small shifts in the proportion of bacteria that are actively synthesizing proteins would result in significantly higher production without necessarily resulting in large increases in respiration rates. This hypothesis remains to be tested.

Estimating Bacterial Carbon Metabolism at the Ecosystem Level

One of the objectives of this study was to determine the importance of spatial variation in estimating the bacterial contribution to ecosystem C flux in a river. Two important assumptions were made in calculating whole ecosystem estimates: that BP and BR did not vary significantly with either depth or from bank to bank in the river. Previous studies have shown that several variables, including BP, do not change significantly with depth in this rapidly mixed, turbulent system (Findlay and others 1991; Cole and others 1992). Preliminary sampling carried out in the month of August at the height of the macrophyte growing season also demonstrated that BP and BR measurements in both *Trapa* and *Valassenaria* weed beds were not significantly higher than average rates in the central channel for the entire of the river (data not shown). In addition, these macrophyte beds represent a small proportion of the entire river in terms of both volume and area. Thus, even slightly higher rates of BP and BR in these specified areas would have little impact on riverwide estimates. However, the latter may not apply to other river systems with more extensive macrophyte coverage. Therefore, the rates of BP and BR measured from surface samples taken in this study were considered spatially representative and sufficient in calculating the ecosystem estimates of BR and BCD.

The lower variability of BR in space implies that less spatial coverage may be necessary for an accurate ecosystem estimate of BR. However, understanding the differences in the fate of bacterial C in a spatial context requires detailed measurements of BP. In the case of the Hudson, as evidenced by the higher average BGE in the northern section of the river, a greater proportion of the BCD is a function of elevated BP in the north relative to the south. Therefore, spatial measurements of BP are important to our understanding of the factors regulating bacterial community dynamics, whereas fewer spatial measures are needed for ecosystem estimates of bacterial C flux, which is primarily driven by BR.

Temporal variation in river respiration rates can be quite significant, so that system-level annual estimates require repeated measurement over time. Temperature appears to be the main factor contributing to this variability because higher observed rates of BR are generally associated with higher temperatures (Roland and Cole 1999; Bouvier and del Giorgio 2002). Respiration rates are apparently also influenced by seasonal and annual variation in discharge (Benner and others 1995; Moran and others 1999; Taylor and others 2003) and likely by the quality of the organic matter associated therewith. Decreases in the rate of respiration have been observed with increasing discharge (Benner and others 1995; Moran and others 1999), possibly as a function of increased DOC concentration coinciding with reduced substrate quality (Moran and others 1999). Interestingly, we found that BR was generally higher with higher rates of discharge in the freshwater portion of the Hudson River. Therefore, in the case of determining the amount C flowing through the bacteria, sampling BR over time is more important than sampling it over space.

Although fewer stations may be required to estimate volumetric BR (and consequently BCD) at the ecosystem level, it is also important to consider variations in river morphology. For the Hudson, the demand for bacterial C per unit volume is considerably different from the demand per unit area (Figure 6A and B). Often, the average depth of the system is used to convert volumetric estimates from a single site to an ecosystem areal estimate. In the case of the Hudson, multiplying the volumetric estimate by the average depth would lead to an underrepresentation of deep sections of the river, as indicated for rkm 68–84 (Figure 6B). Increases in the total C required by the bacteria within the segments of the river primarily followed the accretion of water (Figure 6C) However, the higher rates of bacterial activity in the most upstream section resulted in an eight fold increase in total C demand relative to the most downstream section. If total C demand simply increased as a function of water volume, a factor of 13 would be expected.

Bacterial Respiration in Relation to the Ecosystem Carbon Budget

Approximately 20% of the organic C either entering or produced within the lower Hudson River is respired by the bacteria (Table 1). The system is highly net heterotrophic because respiration substantially exceeds gross primary production (S. Findlay Forthcoming). The areal rate of BR for 2000 was estimated at 171 g C m^{-2} y^{-1} (Table 1). This value may be high given that measured atmospheric flux of CO_2 from the Hudson has previously been estimated at 70–162 g C m^{-2} y^{-1} (Raymond and others 1997) these latter measurements included all heterotrophic respiration, not only that of the bacterioplankton. However, the respiration value we report here is not inconsistent with other published estimates (Cole and Caraco 2001). Thus, the fundamentally important role of bacteria in the mineral transformation of organic C during riverine transport is apparent in the Hudson.

In summary, over one-quarter of the OC entering or produced within the lower Hudson River is either remineralized or converted into biomass by heterotrophic bacteria. The proportion of organic matter C transformed by bacteria in other rivers may be lower (Raymond and Bauer 2000; Raymond and others 2000), but few studies on river ecosystems have attempted to quantify the detailed spatial C budgets presented here. From a spatial perspective, BP varied in a distinct north–south pattern, suggesting that there are important variations in the local factors regulating bacterial growth. Bacterial respiration was more constrained spatially but had important temporal variation. From the point of view of determining the contribution of bacteria to ecosystem C flux, the relative invariance of BR in space is quite convenient: An accurate estimate of bacterial C flux can be determined at a single morphometrically representative station, sampled temporally at least in the case of the Hudson River. However, why BR is such a constrained variable in this and among different aquatic ecosystems remains an open and important question.

ACKNOWLEDGMENTS

R.J.M. was supported by a postdoctoral fellowship from the National Sciences and Engineering Research Council of Canada (MSERC). This work was funded by grants from the Hudson River Foundation and the US National Science Foundation. The paper is a contribution to the Institute of Ecosystem Studies. We thank D. Fisher, G. Lampman, H. Malcom, and A. Nixon for their excellent technical and field assistance; H. S. Canut for guidance; and C. Duarte and two anonymous reviewers for their helpful comments.

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