

Primary Research Paper

Degradation of surface-water dissolved organic matter: influences of DOM chemical characteristics and microbial populations

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Abstract

The degree to which biodegradation of dissolved organic matter (DOM) depends on microbial community structure and source remains unknown. In this study, we concentrated the microbial biomass from two streams in northern Michigan and a dystrophic 'bog' lake in northern Wisconsin with varying initial DOM concentration (6.7–78.8 mg C l⁻¹) and DOM chemical characteristics (e.g. DOM average molecular weights from 808–1887 Da). Each of the three microbial inocula was added to each of the three DOM sources at *in situ* population levels for a total of nine treatments. Changes in DOM concentration and bacterial productivity, along with chemical characteristics, were examined over 308 h. The [³H]-leucine incorporation method was used to measure microbial production. In two of three sampling sites, bacterial communities were most productive when metabolizing DOM in their native waters. A variable peak in productivity was seen between 16–48 h after inoculation, followed by a drop in productivity in most treatments, with periods of DOM production most likely due to microbial turnover. These data suggest that microbial communities are better able to degrade the DOM of their native habitats, suggesting that biodegradation of DOM is influenced by source-specific microbial species and DOM chemical characteristics.

Introduction

Dissolved organic matter (DOM) is a ubiquitous component of surface waters, and it plays an important role in controlling metal and pollutant mobility, attenuating potentially damaging UV radiation, and fueling microbial-based food webs (Findlay et al., 1986; Azam, 1998). Microbial productivity linkages to DOM cycling are of great importance in wetland (Mann & Wetzel, 1995), river and stream (Meyer et al., 1987; Kaplan & Bott, 2000), lacustrine (Tranvik, 1989; Tulong et al., 1992; Bertilsson & Tranvik, 1998), and oceanic environments (Azam, 1998).

DOM in surface-water ecosystems is composed of two broad fractions: the readily utilizable DOM or 'labile pool' and the residue that is 'refractory'

and not as easily utilized (Ogura, 1975; Geller, 1986). Although the definition of labile DOM is somewhat complex, the labile pool is generally thought to consist mainly of sugars, amino acids, peptides and other simple compounds (Moran & Hodson, 1990), and accounts for < 20% of the total DOM. The labile pool turns over rapidly, in a time span of hours to days. Sondergaard & Middelboe (1995) defined labile DOM as the amount of DOM which could be decomposed by bacteria within a week or two; it has been shown to support the majority of bacterial secondary production (Moran & Hodson, 1990). An alternative definition of labile DOM or biodegradable dissolved organic carbon (BDOC) was suggested by Servais et al. (1989), who determined the labile DOM by filter-sterilizing a water sample containing the DOM to be tested,

inoculating it with autochthonous bacteria, and measuring the decrease of [DOM] over four weeks due to carbon oxidation by bacteria.

The refractory pool, which is composed mostly of higher molecular weight humic and fulvic acids, is more abundant, but it turns over more slowly and has been considered relatively less important as a substrate for bacterial growth (Moran & Hodson, 1990) than the labile pool. Nevertheless, at least some components of the refractory pool may be altered photochemically to produce more bioavailable compounds (e.g., Lindell et al., 1995; Wetzel et al., 1995; Moran et al., 2000), and several studies have suggested that humic substances are more important components of the biodegradable DOM pool than previously thought (e.g., Meyer et al., 1987; Amon & Benner, 1996; Volk et al., 1997). Some high-molecular-weight organic compounds appear to be readily utilized by bacteria in both the ocean and in streams (e.g. Tranvik, 1990; Tulonen et al., 1992; Amon & Benner, 1996; Meyer et al., 1997; Volk et al., 1997).

A growing body of evidence suggests that the rate at which bacteria degrade different varieties of organic compounds depends not only on the nature of the carbon compounds, but also on bacterial physiology (Leff & Meyer, 1991). For example, in a study on the Ogeechee River, Leff (2000) found that *Pseudomonas putida*, which is able to use aromatic compounds, increased along the same gradient as observed by Sun et al. (1997) for increase in the DOM aromatic fraction. In a study of a first-order coastal plain stream in New Jersey, Maurice & Leff (2002) found that Domain *Bacteria* cell counts determined from fluorescent *in situ* hybridization (FISH) correlated positively with DOM aromaticity and molecular weight. This suggested that bacteria in the stream were able to use higher molecular-weight, aromatic-rich components of the DOM. Covert & Moran (2001) observed substantial differences in the composition of bacterial communities growing on high molecular weight (> 1000 Da) vs. low molecular weight (< 1000 Da) fractions of an estuarine DOM pool. Therefore, a growing body of evidence suggests that different bacterial communities have different abilities to utilize high- vs. low-molecular weight DOM components.

To provide a direct test of how DOM bioavailability varies with microbial community

structure and quantity and chemical characteristics of DOM, we isolated microbial communities from two streams and a dystrophic lake located in northern Wisconsin and the Upper Peninsula of Michigan and reciprocally inoculated DOM from each site with each microbial community. Our objectives were to determine (i) if DOM bioavailability is a relative concept depending on the microbial assemblages, and (ii) if the same bacteria consortium can metabolize differently depending on DOM chemical characteristics.

Materials and methods

Site description and sampling

Surface water samples were collected from three aquatic sites (two streams and one bog-lake) located in northern Wisconsin and northern Michigan. Two Mile Creek (TMC) and Nelson Creek (NLC) are located in the Ottawa National Forest, and Forest Service Bog (FSB) is located nearby, at the University of Notre Dame Environmental Research Center (UNDERC). FSB, TMC and NLC represent sites of relatively low (6.7 mg C l^{-1}), medium (17.2 mg C l^{-1}) and high (78.8 mg C l^{-1}) [DOC], respectively.

FSB is a small 'bog' lake with an open-water area of $\sim 0.21 \text{ ha}$ and a maximum depth of 4.9 m. The majority of the lake has been filled by an encroaching peat mat of similar maximum depth to the current open-water zone. Small lakes surrounded by peat mats are a common occurrence in this area. The peat mat is vegetated by *Sphagnum* sp. mosses, various sedges, ericaceous shrubs, *Picea mariana* (black spruce), and *Larix laricina* (tamarack) – all typical of peatlands in this region. The relatively low [DOC] and pH of 5.1 (Table 1) in the open-water zone suggest substantial groundwater input from mineral soil in surrounding uplands.

NLC is a first-order stream and TMC a second-order stream. Both streams flow through and/or are surrounded by a number of wetlands, a likely source of their relatively high [DOC]. Their watersheds are predominantly northern hardwood forest (e.g. *Acer saccharum* and *Fagus grandifolia*) in the uplands with both deciduous and evergreen plant species in the extensive wetlands.

Table 1. Physical and chemical characteristics of the study systems – Forest Service Bog (FSB), Twomile Creek (TMC), and Nelson Creek (NLC). DOM molecular weight distributions are of DOM from each site. Standard error in parentheses

	FSB	TMC	NLC
<i>DOM characteristics</i>			
DOC (mg C l ⁻¹)	6.7	17.2	78.8
ϵ_{280} (l mol C ⁻¹ cm ⁻¹)	211	281	368
<i>DOM molecular weight distributions</i>			
M_w (Daltons)	808	1097	1887
M_n (Daltons)	450	669	890
ρ	1.80	1.64	2.12
<i>Inorganic constituents (mg l⁻¹)</i>			
Al	0.016(0.025)	0.016(0.003)	0.433(0.0045)
Fe	0.021(0.024)	0.188(0.0023)	1.455(0.016)
Ca	0.13(0.066)	18.75(0.31)	6.55(0.058)
Mg	Below detection	5.765(0.042)	1.71(0.016)
Na	1.12(0.025)	2.7(0.006)	3.55(0.043)
K	0.281(0.004)	0.575(0.0061)	0.305(0.0018)
<i>In situ measurements</i>			
pH	5.1	7.3	6.2
Conductivity ($\mu\text{s cm}^{-1}$)	9.2	144.5	40.7
Temperature (°C)	17.5	18.6	19.7

Sampling was conducted in early September 2001. Water samples were collected using a polyethylene bottle attached to a pole and filtered on-site, using precombusted glass-fiber Whatman filters (GF/F) and a vacuum filtration system. The samples were stored in acid-washed and autoclaved glass bottles in the dark at 4 °C for DOM preservation. Additional samples to be used for bacterial inoculates were first filtered through 1- μm Nuclepore filters to remove protozoa, although some smaller bacteria likely remained. The samples were stored in the dark at ~ 22 °C. The bacterial inoculate was later re-filtered through 0.22- μm filters during inoculate preparation (see section below). Conductivity, pH, and water temperature were recorded *in situ* and inductively coupled plasma optical emission spectroscopy (ICP-OES) was used to conduct an analysis of inorganic constituent concentrations (Table 1).

Bacterial inocula

The experiment was started within 48 h of collection. DOM samples were moved to a 4 °C incubator and bacterial inoculates were prepared

by centrifugation of 1.2 l from each site using a Beckman J2-HS Centrifuge at 8000 rpm for 30 min at room temperature. Pellets were re-suspended in 0.22 μm filter-sterilized water from the native site, creating a 30-fold concentration of bacteria for each inoculum. Prior to inoculation, each inoculum was visually inspected under the Olympus BH-2 epifluorescence microscope at 1000 \times magnification and no nanoflagellates or protozoans were seen. Three replicate DOM samples from each site (386.6 ml) were inoculated with concentrated bacterial inoculum (13.4 ml) from each of the three sites, yielding 3 replicates of 9 treatments (400 ml each) from each source DOM. This procedure yielded approximately *in situ* population densities of bacteria, and previous data showed that a 100% concentration of inoculate would give optimal results for this procedure (data not shown). As a result of adding the high DOM bacterial inoculum to a low DOM source (i.e. adding the NLC inoculum to the FSB DOM), there was a small increase in the total DOM (Fig. 1). Despite this slight increase in total DOM, there is no indication that the small amount of DOM that may have been transferred

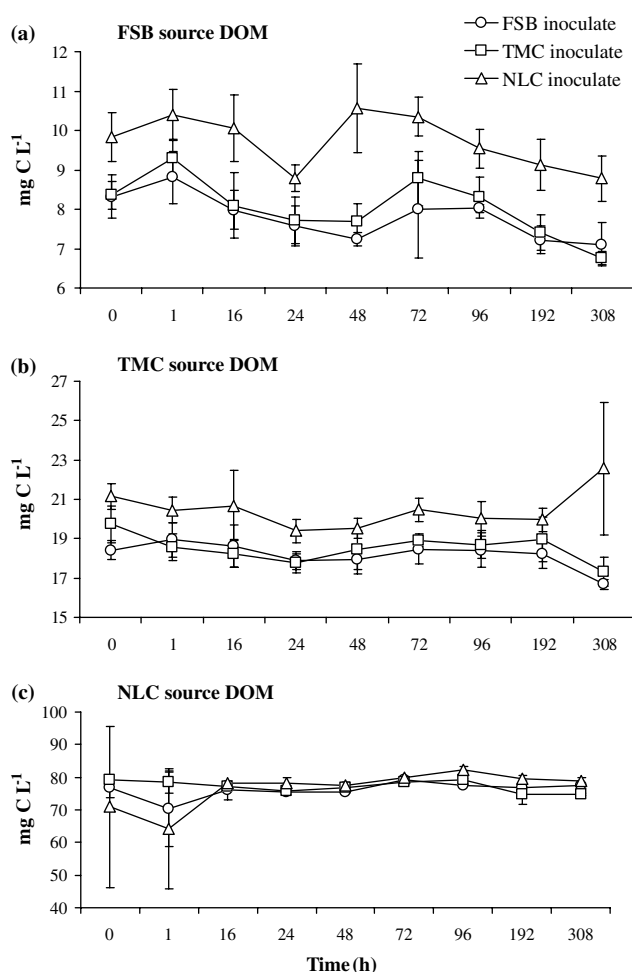


Figure 1. (a) [DOC] in FSB (○), TMC (□), and NLC (△) after each time interval in (a) FSB (low) [DOC], (b) in TMC (middle [DOC]), and (c) in NLC (high [DOC]). Error bars represent \pm SE ($n = 3$). Note differences in y-axis scales.

through the inoculum had any effect on productivity (Fig. 2).

Samples were stored in amber glass bottles at room temperature on a shaker table at 50 rpm. Bottles were stopped with plugs of glass wool to prevent contamination by adventitious bacteria in the laboratory, but to allow for passage of air and to prevent the system from becoming anaerobic.

Microbial growth and cell enumeration

Heterotrophic bacterial biomass production was determined on 5-ml aliquots from each sample at 0, 1, 16, 24, 48, 72, 96, 192 and 308 h by [2,3,4,5-³H] L-leucine incorporation using ³H of specific activity of either 73 Ci mmol⁻¹ or 171 Ci mmol⁻¹. This is a

filtration technique and is explained in detail in Kirchmann (1993). A preliminary experiment that optimized the time and concentration of leucine used indicated that a 1-h incubation with 10 nM ³H-Leu yielded optimal results (data not shown). Counts per minute (CPM) were measured using a Beckman LS 5000 TD and TA Liquid Scintillation System. Disintegrations per minute (DPM) were corrected for quench using an experimentally determined quench curve. DPMs were converted to microbial growth rates using a conversion factor of 3.1 kg C mol⁻¹ (Kirchman, 1993). Productivity numbers were then standardized to per cell values using initial direct microbial counts (see below).

About 5 ml aliquots were removed at 0 h and preserved in 37% formaldehyde for initial cell

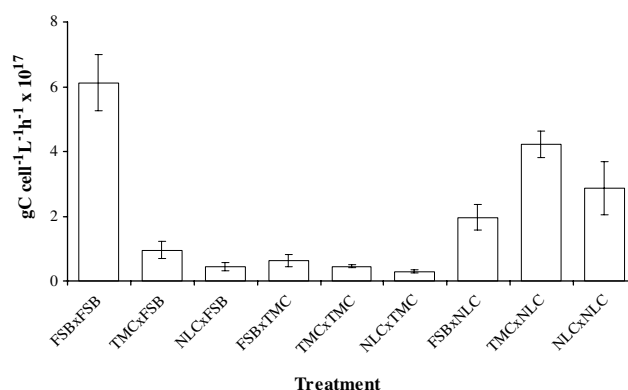


Figure 2. Mean cumulative bacterial productivity over 308 h. Productivity measurements were derived for each time when productivity was actually measured summed for all time points and averaged over 3 replicates. The treatments are statistically different ($p = 0.000$) according to one-way ANOVA. Error bars represent \pm SE of the mean of each replicate for 9 treatments.

density enumeration by direct counts (Hobbie et al., 1977). Two milliliter of each replicate were incubated in the dark with 0.2 ml 0.1% acridine orange (AO) fluorescent stain for 2 min, and then filtered onto a 0.22- μ m Nuclepore filter for direct count analysis. At least 200 cells were counted per slide using an Olympus BH-2 epifluorescence microscope at 1000 \times magnification. Direct bacterial cell counts differed among the three sites (ANOVA, $p = 0.032$) and the range of means for all cell counts was from 4.83×10^7 to 16.2×10^7 . All data have been corrected for total cell counts to remove any variation that might have occurred due to different numbers of bacteria existing in the inoculum. However, relative differences among treatments were similar with or without the correction for initial bacterial cell numbers.

DOM characterization

At each time interval, an additional 5-ml aliquot was removed from each replicate and suctioned through a GF/F-filter for [DOC] analysis. Non-purgeable organic carbon concentrations ([DOC]) were measured by combustion on a Shimadzu TOC5000 with an autosampler. Prior to analysis, samples were acidified with distilled nitric acid and purged for 2 min with CO₂-free air to remove inorganic C. Potassium hydrogen phthalate was used to prepare standards. Samples were run in replicate until the analytical uncertainty, as measured by relative standard deviation, was <2%.

Using the high-pressure size exclusion chromatography (HPSEC) method of Chin et al.

(1994), as revised by Zhou et al. (2000), the molecular weight distributions of DOM samples were measured on samples prior to bacterial inoculation. Briefly, the mobile phase consisted of degassed 0.1 M NaCl buffered with 0.002 M KH₂PO₄ and 0.002 M K₂HPO₄. Waters Protein-Pak 125 modified silica column (30-cm long, 7.8-mm diameter) was used with the Waters HPSEC instrumentation (Waters Associates, Milford, MA). All standards and samples were detected at 254 nm. It should be noted that small molecules are often invisible to the UV-Vis detector used in HPSEC determinations (e.g., O'Loughlin & Chin 2001); therefore, the average molecular weights reported herein represent primarily humic and fulvic acids and are biased against the low-molecular weight components (particularly, < 50 Da) of the DOM pool. Using the equations presented in Chin et al. (1994) and the chromatograph processing procedures described by Zhou et al. (2000), weight-average molecular weight (M_w), number average molecular weight (M_n), and polydispersity (ρ) were determined. M_w is the weight of the molecule to which the 'average' atom belongs. M_n is the weight of the 'average' molecule in the mixture. In a pure substance, M_w is equal to M_n . For a mixture of molecules, $M_n < M_w$, so that $\rho > 1$ (Zhou et al., 2000).

A double-beam Varian Cary III scanning spectrophotometer was used to measure absorbance at 280 nm. One-centimeter quartz cells were used, and Milli-Q water was used as a reference. Molar absorption coefficients (ϵ_{280}) were calculated based on absorbance at 280 nm and [DOC] measurements

($\epsilon_{280} = A_{280} [\text{DOC}]^{-1}$, with units $\text{l mol C}^{-1} \text{ cm}^{-1}$) of organic matter in solution (e.g., Traina et al., 1990; Chin et al., 1994). This technique is useful because 280 nm is the region of π to π^* transitions, so that absorbance at this wavelength is sensitive to the aromatic nature of the DOM.

Statistical analyses

Repeated measures Analysis of Variance (rmANOVA) was conducted for bacterial productivity and change in [DOC] over time, using inoculate source and DOM source as independent factors. All data were tested for a normal distribution using a Lilliefors test, and the productivity data were normalized using a log transformation. Post hoc Tukey's HSD tests were performed on all factors having a significant rmANOVA effect. All analyses were performed using Systat 10, and all significance levels were evaluated at $\alpha = 0.05$.

Results and discussion

Site chemical characteristics

FSB had the lowest [DOC] (6.7 mg C l^{-1}), NLC the highest [DOC] (78.8 mg C l^{-1}) and TMC intermediate [DOC] (17.2 mg C l^{-1}). The pH values of the three sites ranged from mildly acidic to circum-neutral (Table 1). Conductivity, pH, and concentrations of Ca^{2+} , Mg^{2+} , and K^+ were highest in TMC, lowest in FSB, and intermediate in TMC. NLC had significantly higher Fe and Al concentrations than the other two sites, most likely because of complexation by abundant DOM.

The initial (unreacted) FSB, TMC, and NLC DOM samples eluted from the HPSEC column as broad, monomodal distributions (Fig. 3). In HPSEC analysis, lower molecular weight molecules have longer retention times because they take a more tortuous route through the micropores of the column. Thus, the average molecular weight of DOM increased from FSB (808 Da) to TMC (1097 Da) to NLC (1887 Da). Thus, the high [DOC] site, NLC, had the highest M_w , M_n , and ϵ_{280} , whereas the lowest [DOC] site, FSB, had the lowest M_w , M_n , and ϵ_{280} (Table 1).

Experimental considerations

In this experiment, one of the main objectives was to preserve the DOM as unaltered as possible. Therefore, of necessity, there were a number of environmental variables that were left uncontrolled that may influence microbial consumption of DOM. The pH and ionic strength of each treatment were not controlled in this experiment and yet could potentially affect bacterial productivity. Altering the pH by adding HCl or NaOH or changing the ionic strength with an electrolyte solution may have impacted the microbial community and/or changed DOM chemical characteristics more than it was deemed desirable for this experiment. For example, Zhou et al. (2001) observed a decrease of M_w 2290 to 2250 to 2130 as pH decreased from 7.5 to 5.5 to 3.5 for an aquatic fulvic acid. The experiment was also designed to preserve the bacterial pellets in the inoculum as unaltered as possible. Each pellet was resuspended into $0.22 \mu\text{m}$ -filtered water from each bacteria's native site in order to keep cells intact, as

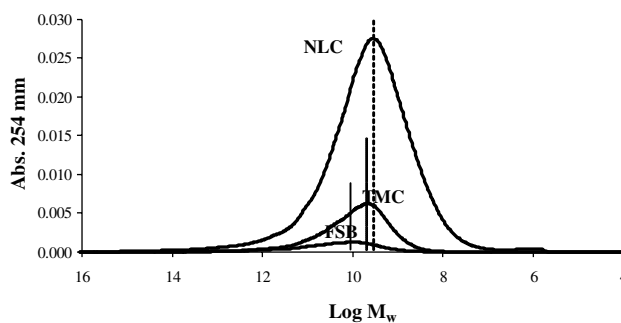


Figure 3. Comparison of HPSEC chromatograms from surface waters at FSB, TMC, and NLC. Vertical lines indicate peak M_w for each DOM sample; line length is irrelevant.

resuspension in a neutral solution such as DI water may have caused bacterial cells to rupture.

Reche et al. (1998) suggested that reducing inorganic nutrient limitation would promote bacterial growth. Although nutrient concentrations were not measured during this experiment, N and P were monitored during the first 72 h of a subsequent but similar experiment using surface waters and bacteria from the same sites (unpublished data). In the subsequent experiment, neither N nor P were found to change significantly over time.

Productivity

Both FSB and TMC were able to grow substantially and to consume DOM even in their non-native waters despite differences in pH, ionic concentration, and different DOM average molecular weight. NLC consumed DOM slightly in the non-native water. Bacteria from both FSB (pH = 5.1) and TMC (pH = 7.3) were able to grow substantially in DOM from NLC (pH = 6.2). Bacteria from the site with the lowest pH, FSB, were even able to survive, grow, and consume DOM in the DOM from the site with the highest pH, TMC (Fig. 4).

Both inoculate source and DOM source had significant effects on microbial productivity (rmANOVA, $p < 0.001$), as well as a significant interaction effect ($p = 0.009$) with time ($p < 0.05$). The effect of time is evident graphically (Fig. 4). For most treatments, there was a lag time in growth between 1 and 16 h, followed by a period of increased growth, with a maximum near 24 h. Thereafter, bacterial growth dropped off rapidly, so that by 72 h it was similar to initial low values in most treatments. FSB bacteria in their native DOM source showed a smaller decrease in growth after 24 h. After 72 h, bacterial growth remained low until 308 h in TMC and NLC inoculates in FSB-source DOM and in all inoculates in NLC-source DOM. However, microbial growth increased again after 72 h in TMC-source DOM, but absolute rates were always very low no matter the inoculate source. The exception to these trends was the FSB inoculate in FSB-source DOM, which maintained relatively high growth rates through 192 h, before decreasing at 308 h. Also, NLC bacteria maintained very low and almost invariant growth in all but their native DOM source.

The period of high bacterial growth from approximately 16 to 48 h was accompanied by an increased rate of DOM consumption (i.e. a noticeable decrease in [DOC]), probably of the most labile fraction of available DOM (Fig. 1). In FSB DOM, there was an average decrease in [DOC] of 9.0% from 0 to 24 h, whereas in TMC DOM, there was an average decrease of 6.9% in the same time period. The change is less noticeable in the NLC DOM. Thereafter, there were periods of production and consumption of net DOM.

The decline in bacterial productivity after 24 h (Fig. 4) was likely due to exhaustion of the most readily available, labile DOM pool, although other factors such as accumulation of toxic byproducts could also be important. Periods of net DOM production after 24–48 h (Fig. 1) suggest turnover of the microbial population, cell lysis, and/or production of labile DOM, which would fuel the secondary peaks in microbial productivity seen in many of the treatments. This is unlikely a reflection of predation because protozoa had been removed by filtration, and microscopically inspected initial samples showed no sign of protozoa. The clearest example of this secondary peak was with FSB bacteria in the FSB-source DOM. However, it is possible that there was a succession in the microbial communities over time, with selection for those populations more capable of degrading the remaining, more refractory DOM. Massana et al. (2001) examined initial and final microbial assemblages in long-term incubations and found a significant change in species composition when incubated without predators. They concluded that all microbial populations in long-term incubations do not grow harmonically. In our data, the secondary peak in microbial productivity is likely caused by activity of a few opportunistic bacteria, rather than the entire original microbial assemblage. Despite this, while DOM is naturally and continually produced in natural aquatic ecosystems from autochthonous and allochthonous sources, our data suggest that labile DOM is recycled over days to possibly weeks due to turnover of microbial populations.

The highest maximal peak of bacterial growth at 24 h ($> 3.0 \times 10^{-17} \text{ g C l}^{-1} \text{ cell}^{-1} \text{ h}^{-1}$) occurred in FSB bacteria in FSB-source DOM and in TMC and NLC bacteria in TMC- and NLC-source

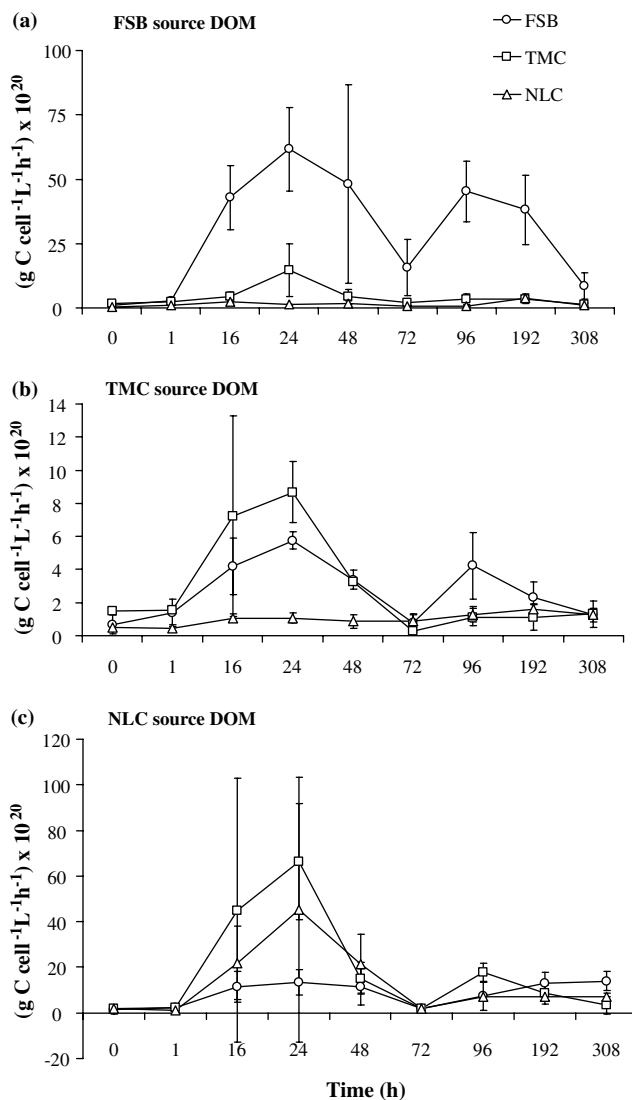


Figure 4. Mean productivity FSB (○), TMC (□) and NLC (△) bacteria in (a) FSB (low initial [DOC]), (b) in TMC (middle [DOC]), and (c) in NLC (high [DOC]). Error bars represent \pm SE ($n = 3$). Note differences in y-axis scales.

DOM. The [DOC] (initial and at 24 h), the DOM average M_w , and ϵ_{280} exerted at most only a small influence in bacterial productivity at 24 h ($R^2 = 0.4306$ for initial [DOC], 0.4360 for 24 h [DOC], 0.3768 for M_w , and 0.280 for ϵ_{280} , all positively correlated). We did not perform multiple regression analysis because the [DOC] and DOM properties have a high degree of autocorrelation. This suggests that a substantial proportion of the variability in microbial productivity at 24 h is likely to be explained by variations in

microbial community structure, or possibly due to other unmeasured environmental factors.

Cumulative bacterial productivity over the 308 h incubation was highest with FSB bacteria in FSB-source DOM and in all 3 inoculum sources in NLC-source DOM (Fig. 2, ANOVA, $p < 0.001$). The high bacterial productivity with NLC DOM, which had both the highest [DOC] and also the most recalcitrant carbon on average as determined by M_w , M_n , and ϵ_{280} of the three DOM sources (Table 1), suggests that the absolute quantity of

DOM exerts an important influence on bacterial growth.

However, other factors are clearly important in determining bacterial growth, as FSB bacteria in FSB-source DOM had the highest cumulative productivity, despite having low [DOC]. Additionally, FSB bacteria exhibited higher metabolic activity in FSB-source DOM than TMC and NLC bacteria at every time interval following 1 h (Fig. 4a). Thus, this site had the lowest average molecular weight and least aromatic DOM as determined by M_w , M_n , and ϵ_{280} (Table 1), but other bacterial inoculates did not have high productivity with this DOM source. TMC bacteria also exhibited higher metabolic activity in TMC-source DOM than FSB and NLC bacteria at their period of peak activity at 16 and 24 h (Fig. 4b), although this difference was not evident in the cumulative data (Fig. 2). NLC bacteria also had a high metabolic rate in their native DOM source (Figs. 4c and 6), although TMC and NLC bacteria were not significantly different.

Our data suggest that that DOM quantity and chemical characteristics may be important in determining microbial growth rates, but that the relative importance of these two factors depends on microbial community composition. For example, Cottrell & Kirchman, (2000) found that different bacterial phylogenetic groups are dominant in different basic types or ‘qualities’ of oceanic organic matter samples. *Cytophaga-Flavobacter* is most representative in recalcitrant DOM, while α -Proteobacteria is most representative in labile sources. Covert & Moran, (2001) used a variety of techniques, including sequencing and terminal restriction fragment length polymorphism (TRFLP) analysis of 16S rRNA gene amplicons to investigate the compositions of bacterial communities growing in high vs. low molecular weight fractions of estuarine dissolved organic carbon. The results of two different studies conducted at the same site revealed differences in communities utilizing the high vs. low molecular weight fractions, which the authors suggested could be due to chemical differences in the dissolved organic carbon components. Although our study likely would have benefited from the use of molecular microbial community profiling, the preponderance of our results supports our second hypothesis that native

bacterial communities are selected to have optimal growth with their native DOM sources. This study provides new insight into microbial–DOM interactions, but in future work, community level molecular approaches such as denaturing gradient gel electrophoresis (DGGE) or TRFLP would likely provide additional causal information on the link between structure and function in microbial–DOM interactions.

Much research to date has shown that lower molecular weight fractions of DOM are the most bioavailable for uptake (e.g. Saunders, 1976; Hobbie, 1988; Meyer et al., 1997; and references therein). Contrary to this, several studies have shown that higher molecular weight fractions are readily taken up by bacteria in marine and fresh waters (e.g. Tranvik 1990; Tulong et al. 1992; Amon & Benner, 1996; Meyer et al., 1997; Volk et al., 1997). Our study did not address low-molecular weight components which were not accessible by HPSEC. However, we observed that the lowest M_w DOM sample, FSB, was only bioavailable to the FSB community. The highest M_w DOM, NLC, was bioavailable to NLC, TMC and, to a small extent, FSB communities. Although other parameters (e.g. pH, ionic strength) were not held constant, these observations suggest that average molecular weight is not the primary factor that controls bioavailability of the fulvic-type DOM components.

Conclusions

Our data suggest that that DOM quantity and chemical characteristics may be important in determining microbial growth rates, but that the relative importance of these two factors depends on microbial community composition. In the majority of cases microbial communities grew best on their native DOM source. Additionally, there was significant production of labile DOM after several days, likely from turnover of the microbial population, suggesting repeated recycling of labile DOM in aquatic ecosystems. This study demonstrates that a better understanding of microbial community structure is essential for prediction of DOM dynamics in aquatic ecosystems and, hence for proper engineering of DOM-sensitive water treatment issues.

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